









# PHYSIOLOGICAL CHEMISTRY

BY

PROFESSOR C. G. LEHMANN.

TRANSLATED FROM THE SECOND EDITION

BY

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EDITED BY

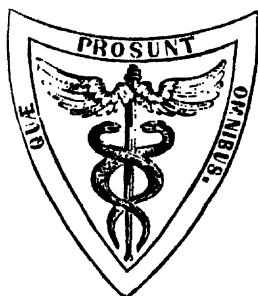
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WITH ILLUSTRATIONS,  
SELECTED FROM FUNKE'S ATLAS OF PHYSIOLOGICAL CHEMISTRY,  
AND  
AN APPENDIX OF PLATES.

COMPLETE IN TWO VOLUMES.

VOL. I.



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## PREFACE OF THE AMERICAN EDITOR.

THE present edition of Lehmann's "Physiological Chemistry," is a reprint of the translation by Dr. G. E. Day, of London, from the second edition of the German.

In undertaking to superintend this valuable work in its passage through the press, I have felt that by contributing to give it a wider circulation than was consistent with the limited edition of the "Cavendish Society," I would be rendering a service to those interested in the subjects of which it treats.

As respects my execution of the task, I would desire it to be understood, that I have confined myself to the careful distribution throughout the sections of each volume, of the considerable amount of matter contained in the "Appendix," and to a critical revision of the proof-sheets; and that in no instance have I introduced any important alterations, either in the text of the author or in the notes of the translator. Occasionally, in the process of interpolating the additions of the Appendix in their proper places, it became necessary to change or omit a word or even an expression, for the sake of preserving the connection between the portions of the work which had been written earlier, and those more recent observations and discoveries which the author and translator were unable to introduce in the body of the text; but in no case has this freedom been exercised except where it appeared indispensable.

To give additional value to the work, a selection of illustrations has been made from the beautiful Atlas of Dr. Otto Funke, which it is hoped will be found of service as a guide to the student of medical microscopy, in his examinations of the substances to which they refer.

With the same view, there have been added at the end of the work, and in a separate form, a number of wood-cuts, selected from various works on allied subjects, which may be usefully consulted by the reader

during his study of the text, or by furnishing in compact form the means of convenient comparison, may perhaps be of service to those engaged in original research.

The high reputation of Professor Lehmann as a chemist and experimental physiologist, the known competency of the translator, and the established character of the "Cavendish Society," under whose auspices it has appeared in England, furnish an ample guarantee of the pre-eminent value of this work, as well in regard to its practical details as its speculative inquiries.

These testimonials to its excellence are sufficient to commend it to the student as the most accurate and thorough work of reference on the subject of Zoo-Chemistry, to the medical practitioner as a valuable guide in the solution of many of the difficult questions in which he is interested, and to the experimental inquirer who, in his endeavors to extend the boundaries of knowledge, feels the want of such a critical and complete exposition of what has already been determined.

R. E. R.

GIRARD STREET, PHILADELPHIA,

OCTOBER, 1855.

## TRANSLATOR'S PREFACE.

IN presenting Lehmann's "Physiological Chemistry" to the Members of the Cavendish Society, I feel that it would be superfluous to offer any remarks on the author's high reputation as a general cultivator of chemical science; to recapitulate his numerous and important contributions to physiological chemistry; or to refer to the very favorable reception which this work has received in Germany.

The first edition of this volume appeared in 1841, and the second (from which this translation is executed) in the beginning of last year.<sup>1</sup> If, during that interval, the progress of physiological chemistry has been so rapid as to necessitate the entire remodelling of the work (see p. vii), the shorter period that has elapsed since the appearance of the second edition has been proportionally fruitful in important discoveries. Need I advert to the detection of succinic acid as a morbid product in the human organism, to the later researches of Schwartz on hippuric acid and the hippurates, to the detection of hippuric acid in the blood of the ox, and of oxalic acid in diseased human blood, to the discovery of hypoxanthine and inosite, or to Liebig's important memoir on the fibrin of muscular fibre?

As Professor Lehmann will probably append a supplement to his third and concluding volume, so as to embrace a notice of the discoveries which have been made during the progress of publication, I have abstained from anything beyond the very briefest enunciation of any of these recently discovered facts, and have frequently contented myself with a mere reference to the original source of information.

I have deemed it advisable not to interfere with the thermometric scale, weights, and measures, that are now almost universally adopted

<sup>1</sup> Lehrbuch der physiologischen Chemie. Von Prof. Dr. C. G. Lehmann. Erster Band. Zweite gänzlich neu umgearbeitete Auflage. Leipzig, Verlag von Wilhelm Engelmann. 1850.

on the Continent. Degrees of temperature in this work are always expressed in the centigrade scale, but at page xiv, the reader will find a table by which he can, at a glance, discover the degrees, according to Fahrenheit, corresponding with every temperature referred to in this volume. The gramme has now become a recognized standard weight in all our laboratories; in all the cases where it occurs in this work, sufficient accuracy will be attained if we regard it as equal to fifteen grains and a half.

The author, in his foot-notes, very commonly refers to German translations or abstracts of French and English memoirs; in almost every case I have given the corresponding reference to the original source. His numerous references to Dr. Golding Bird's researches are made to Eckstein's translation of a Course of Lectures by that gentleman, which appeared nine years ago in the "Medical Gazette," and I have deemed it expedient slightly to modify a few sentences in the text, which express views somewhat different from those given in the third edition of the "Urinary Deposits."

If, in a few cases, I have ventured to deviate from the ordinary nomenclature,<sup>1</sup> I have not done so without due consideration, and without the sanction of the most competent judges.

I cannot allow these pages to leave my hands without expressing my general obligation to the Council of the Cavendish Society for the readiness with which they accepted my suggestion, that a translation of Lehmann's "Physiological Chemistry" should appear under their auspices, and for intrusting me with the office of Editor. To Professor Graham, Dr. Hofmann, Mr. Redwood, and Dr. Pereira, I am specially indebted, for much kind aid and many valuable suggestions.

G. E. D.

ST. ANDREWS,  
July 9th, 1851.

<sup>1</sup> I have, as a general rule, adopted the final syllable, *ine*, both for the true alkaloids, and for those allied substances which are described in the same section, but do not present any very distinct basic characters, as, for example, *creatine*, *allantoine*, and *cystine*. The terminal *in* refers to neutral bodies, as, for instance, *asparagin*. I have felt considerable difficulty in the nomenclature of the acids: most commonly I have converted the German antepenultimate *in* into *ic*; thus, *Inosinsäure* is translated *Inosic acid*; *Vaccinsäure*, *vaccic acid*, &c.

## AUTHOR'S PREFACE.

SINCE the publication of the first edition of this work, Chemistry—and more especially Physiological Chemistry—has been so zealously and extensively cultivated, and has been enriched by the acquisition of so large a mass of new facts and discoveries, that we may regard the last ten years as one of the most important periods in the history of this science. Hence a simple enlargement of the early edition would not have enabled us to consider all the advances made within this short period, which rather required that the whole work should be entirely remodelled, both in relation to its form and contents. The most superficial comparison of the two editions will suffice to show that this volume has been subjected to so entirely new a mode of arrangement, that only a few paragraphs have been borrowed from the earlier edition; for thus alone could a faithful representation of the present state of this department of chemistry be afforded.

The rapid advance of science and the extraordinary accumulation of a mass of crude materials, some of which may not even be capable of acquiring form, must plead in extenuation of the delay that has attended the publication of the second volume. There are, however, two causes which render this delay in some degree pardonable. The one depends upon the intimate connection of the objects under consideration with histology, the history of development, and pathological anatomy; and as the censure, which has more or less justly been thrown on the writers on physiological chemistry, may be traced to ignorance or neglect of the kindred branches of science, the author has endeavored to fit himself for the task of critically reviewing the labors of others, by acquainting himself, through personal observation and experience, with the grounds on which these departments of science are based. The great mass of voluminous and often obscure materials presented by physiological and pathological histology must necessarily be subjected



to a critical examination before they can be incorporated with physiological chemistry, and hence the author regards such a course of self-training as indispensable in the attempt to furnish his readers with a systematic arrangement of facts. Moreover, those departments of science which must serve as a basis to physiological chemistry, have been encumbered with an accumulated mass of observations, from which have arisen numerous hypotheses successively displaced by others not unfrequently of an opposite character. We must, therefore, as far as is possible, attempt to judge for ourselves if we would not be continually drawn aside by the opinions which are ever rising and falling amid the fluctuations of ephemeral literature.

But the most important reason for the delay that has occurred in the publication of the second volume is, that in Physiological Chemistry, even more than in Zoo-Chemistry, we are obliged to depart from the sure ground of exact inquiry, and to proceed to the consideration of chemico-vital processes, which lie beyond the scope of direct observation, and are thus called upon to admit the correctness of deductions, whose logical authority is not always easy of recognition. Modern science has directed its highest energies to this point of physiologico-chemical investigation; and it was therefore to be expected that this yet imperfectly cultivated soil would give birth to a number of more or less ingenious hypotheses, which can only be sifted by independent examination and positive investigations. But since even this protracted delay and the frequent reconsideration of all the materials at his command do not give as satisfactory a result as the author could wish, he has at length determined to send forth this attempt at a History of Physiological Chemistry, trusting to the indulgence of those who are laboring in the same cause.

LEIPSIC,

September, 1840.

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C.	F.	C.	F.	C.	F.	C.	F.
—123° =	—171·4°	44° =	111·2°	85° =	185°	155° =	311°
— 20	— 4	49	120·2	90	194	157	314·6
— 15	+	50	122	92	197·6	160	320
— 9	15·8	55	131	99	210·2	165	329
— 1	30·2	56	132·8	100	212	170	338
0	32	56·3	133·3	105	221	176	348·8
4	39·2	56·5	133·7	106	222·8	178	352·4
6	42·8	57	134·6	107	224·6	180	356
7	44·6	58	136·4	110	230	182	359·6
10	50	60	140	115	239	195	383
14	57·2	61	141·8	116	240·8	200	392
15	59	62	143·6	117·3	243·1	202	395·6
16	60·8	63	145·4	118·5	245·3	205	401
17·5	63·5	64	147·2	120	248	210	410
20	68	65	149	125	257	215	419
25	77	65·5	149·9	127	260·6	220	428
26	78·8	68	154·4	130	266	228	442·4
30	86	70	158	133	271·4	232	449·6
32	89·6	73	163·4	135	275	236	456·8
35	95	75	167	136	276·8	239	462·2
36	96·8	76	168·8	137	278·6	240	464
37	98·6	78	172·4	140	284	250	482
38	100·4	79	174·2	145	293	255	491
40	104	80	176	150	302	300	572
42	107·6	83	181·4	152	305·6	360	680

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## NOTE.

Page 197, last line, "he never operated on less than two pounds of blood." Lehmann has here fallen into the error of mistaking grains for grammes. Garrod states, in his Memoir in the 31st volume of the *Medico-Chirurgical Transactions*, that he usually employed 1000 grains of serum, which is equivalent to about two fluid ounces. I am assured by him, that in no instance has he caused the abstraction of more than six ounces of blood from gouty patients, and in most cases the quantity did not exceed three or four ounces.—G. E. D.

# PHYSIOLOGICAL CHEMISTRY.

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## METHODOLOGICAL INTRODUCTION.

THE application of Chemistry to the elucidation of physiological and pathological processes has been so universally admitted during the last ten years, that it would appear almost superfluous to commence this work with any observations on the importance of this science. While at no very remote period we had occasion to defend this recent department of chemical science from the attacks and unfavorable criticisms called forth by its injudicious application, and by the numerous misconceptions which characterized its early development, we are now almost constrained to withhold from it the confidence which has been too liberally awarded it. Enthusiasm in the cause of organic chemistry has degenerated amongst many physiologists and physicians into a fanaticism, which, even in the best cause, tends to invalidate a host of truths in its endeavors to uphold some single fact. We might be disposed to ask, whether its most zealous partisans have not retarded rather than accelerated the period at which it will attain its proper share of appreciation, and its just recognition. In commencing, therefore, the subject of physiological chemistry, nothing is more important than clearly to understand the nature of the results which this department of science is now capable of yielding, and the requirements which, in its present stage of development, it fulfils; and to ascertain the course, the means, and the methods most likely to lead us safely within its domain, and at the same time the best adapted to promote its further progress.

In entering upon this subject, it may not be altogether unprofitable to begin by indicating the numerous errors into which those most zealous in their endeavors to elucidate physiology and medicine, have occasionally been led by chemical theories and inquiries. These errors appear to us to have diverged in three different directions. In the first place, too little attention has been directed to the laws of a true natural philosophy, whose simplest rules have in many cases been wholly disregarded; in the next place, the necessary causal connection existing between chemistry and physiology, as well as between histology and pathological anatomy, has too often been entirely neglected; and lastly, much mis-

conception has arisen from the assumption that chemistry afforded a satisfactory solution to many questions which it is either wholly incompetent to answer, or which must at all events remain undecided in the present state of our knowledge.

While we still find occasion to deplore the absence of the steady influence of a true natural philosophy in the application of chemistry to the science of general life, we do not refer to any of those nearly exploded systems of natural science which may be regarded almost in the light of poetic fictions, but to that Newtonian method of contemplating nature, which has carried Astronomy to its present high state of perfection, and has led to the most brilliant discoveries in physics. It is this method of viewing nature which Fries alone understood how to raise into a system, and to which the immortal Humboldt has given life and expression in his "Cosmos." It is only by the application of abstract physical laws, by the establishment of certain momenta of empirically observed phenomena, and by a steady adherence to safely guiding maxims,—in short, by logical sequence,—that we can advance in the investigation of vital phenomena. It would almost seem as if medicine, in the earlier periods of its history, had cast a shadow over those kindred sciences which are able to afford it aid and support, clouding even their brightest points. It has thus been found impracticable at once to rid medicine, notwithstanding its assumed physiological character, of the mania of attempting to explain everything by the old system of hypotheses; and hence this science has derived less benefit than many others from the exact method of physical inquiry, having simply borrowed certain materials from chemistry and the kindred natural sciences, and substituted, in the place of the older vagaries of natural philosophy, various chemical phrases and high-sounding terms, scarcely less devoid of true import than the former. This deficiency in logical sequence, which we so frequently at present encounter in medicine, has unfortunately also infected animal chemistry; for here likewise facts have not been sufficiently distinguished from hypotheses, or hypothesis from fiction. This is more easily accounted for in physiological than in pure general chemistry: for while the latter treats almost exclusively of palpable phenomena and of well-established facts, which easily admit of being reduced to definite laws, in the former we must necessarily have recourse to experiments and natural investigations, whose success must in a great measure depend on individual operations of the mind. Zoo-chemical processes are the most complicated of any comprised in the domain of natural inquiry; but such processes are not capable of tangible demonstration, but must be divined, or rather, intellectually apprehended. Our senses are incapable of perceiving the causal connection of things, or the logical succession of phenomena; thus we do not see motion, but simply recognize it by the result of the changes effected by it; we do not perceive heat, but simply the variations of the temperature, and the results to which they give rise, &c. Hence it is not our senses which here deceive us, but the judgment which we form regarding the objects presented to us by the perceptive faculties. The causal connection of several allied phenomena (*i. e.*, a process), can therefore only be comprehended by the subjective combination of individual objects

perceived by the senses, and not by sensuous intuition alone. But as soon as we subject to investigation the highly complicated chemical phenomena of life, we enter upon the actual domain of hypothesis. It unfortunately happens, however, that the correct logical conception of an hypothesis has been completely lost sight of, and its place supplied by the vaguest fictions; whence the term has fallen into such discredit that many have been desirous of setting aside all hypotheses, unmindful that even the simplest form of experiment cannot be prosecuted without their aid. Hypotheses are indispensable in every physical inquiry, and must constitute the base of every experiment, as they are in fact merely the subjection of our thoughts and mode of intuition to the reality of phenomena. The question, however, always is, whether the facts at our command logically justify such a procedure, since where such is not the case, the deduction at which we arrive is undeserving the name of an hypothesis, and is a mere fiction, supported at best on a hypothetical foundation.

Physiological chemistry has given rise to many delusions of this nature, owing to its imperfect development, and to the necessity presented by physiology and pathology for chemical elucidation. Some few isolated deductions were drawn from superficial chemical experiments, and arranged in a purely imaginary connection by the aid of chemical symbols and formulæ, for whose establishment analysis in many cases did not even afford any sanction. Thus, for instance, in the attempt to form a conclusion regarding the metamorphosis of the blood from an elementary analysis of its solid residue and of the composition of the individual constituents of the excretions, there is an utter absence of all scientific groundwork; for, independently of the fact that the elementary analysis of so compound a matter as the blood is incapable of yielding any reliable results, and cannot, therefore, justify the adoption of any special chemical formula, it is assuredly most illogical to attempt to compare the composition of the blood collectively, with that of the separate excrementitious matters. In such deductions, expressed by chemical formulæ, the addition of atoms of oxygen, and the subtraction of those of water, carbonic acid, and ammonia are wholly arbitrary: for chemical analyses do not afford the slightest grounds for the majority of these equations. When, on the other hand, we have seen uric acid decomposed by different oxidizing agents into urea and other bodies, and when, further, we find the quantity of uric acid increased in the urine in those cases where a diminished quantity of oxygen is proved to be contained in the blood, we are justified in concluding that also in the animal organism a portion, at least, of the urea found in the urine must have been produced by the oxidation of the uric acid. In the formula which expresses this deduction, we have an hypothesis, but a well-grounded one, which, although requiring further confirmation, is yet wholly different from the frequently condemned, but rarely avoided, abuse of chemical symbols. Chemical equations having no other foundation than the presumed infallibility of empirical formulæ, must, however, cause us to deviate from the path of physical inquiry, and involve us in a chaos of the most untenable delusions. Thus, for instance, a chemical equation might lead us to conclude that glycine (glycocoll) was the source of urea

and lactic acid in the metamorphosis of the animal tissues; for we might conclude that 2 equivalents of hydrate of glycine were decomposed into the above-named substances according to the formula,  $C_8H_{10}N_2O_8 = C_2H_4N_2O_2 + C_6H_6O_5.HO$ . All experiments hitherto instituted with glycine are, nevertheless, opposed to such a disintegration. If, then, we would deduce urea and lactic acid from glycine, which has not been proved to exist in the blood, we should be neglecting the most comprehensive rule of logic, according to which one hypothesis cannot be supported by another. It has, however, unfortunately been too much the practice in recent times to employ far more complicated equations as supports for such purely subjective modes of contemplation, by which a semblance of the most exact method of investigation has been assumed. By these means a number of chemical fictions have supplanted the fancies of that speculative natural philosophy which in earlier times encumbered the study of physiology and pathology, and have plunged medicine into the midst of a new labyrinth of untenable theories.

We have indicated a further cause of the partial failure of the application of chemistry to vital phenomena, in the imperfect causal connection among the different branches of natural science, without which there can be no proper insight into the course of different phenomena, or any recognition of the complete vital process. This is especially the case in reference to pathologico-chemical inquiries, in the majority of which the data yielded by pathological anatomy, and the diagnosis thus afforded, have been too little regarded, whilst the adherents of the pathologico-anatomical school have made free use of chemical phrases and fictions, without an adequate acquaintance with the general science of chemistry. If chemical investigations regarding objects belonging to pathological anatomy would aspire to a scientific value, and if they are to afford any true elucidation of pathological processes, it will assuredly be admitted that the question should be adequately considered from an anatomical and diagnostic point of view. Yet every day presents us with instances of the most flagrant neglect of this self-evident proposition. How frequently we hear of the chemical examination of diseased bones, without any regard to a diagnosis at all in accordance with the present condition of pathological anatomy! What numerous analyses have been made of the bones in osteomalacia, notwithstanding that the morbid appearances of these bones vary so much as to render a definite diagnosis a matter of extreme difficulty to the pathological anatomist! We even more frequently meet with similar inconsistencies in the investigation of diseased animal fluids. Here, as in the statistical method of observing diseases, none but the simplest form of a disease should be made the subject of such inquiries. Yet the casual results yielded by an examination of the urine and the blood in the most complicated forms of disease, are frequently made the sole grounds for drawing conclusions regarding the morbid process itself. In many cases even the true diagnosis of the disease has not been given. Thus, for instance, we are told that the blood has been analyzed in typhoid pneumonia, yet when we read the history of the case, we find that the disease was neither ordinary abdominal typhus with pneumonic exudations, nor what is termed pneumo-typhus, but simple pneumonia with cerebral symptoms.

More frequently still, we are obliged to rest content with vague names of disease, unsupported by any history of the case. In most cases certainly the name of the disease is unimportant. It is by no means essential to the scientific comprehension of such inquiries that the whole history of the case from beginning to end should be given with the circumstantiality at present so much in requisition; but we undoubtedly ought to indicate the condition of the patient, as ascertained by a physical examination, at the period of the removal of any morbid product for chemical investigation. It is the practice in reporting chemical investigations, to detail as minutely as possible the method pursued, that the reader may be able to judge for himself, and test the correctness of each individual step. A similar rule should be observed with reference to the state of the disease in all pathologico-chemical investigations, for it is only by these means that we can impart scientific value to such inquiries. We shall find, however, on examining our pathologico-chemical literature, that this principle is too frequently neglected.

If we would render chemistry truly useful to other departments of natural science, we must be careful to acquire a proper knowledge and a due estimate of the advances made in each; a point which has unfortunately been too much disregarded in reference to histology. We have passed the age when morbid tumors, without regard to their histological constitution, were crushed and pounded in a mortar, with the view of extracting from this artificially produced chaotic mass a principle peculiar to cancer or pus,—a scirrhus or a pyin; but at the present day the combustion tube is still misused in the determination of the elementary composition of a mass made up of the most heterogeneous organic parts. Such analyses are wholly devoid of chemical or physiological value, and cannot, as all chemists must allow, in any way contribute to extend the domain of chemistry, while they are useless alike to the physiologist and the pathologist, being utterly devoid of all scientific links of connection. If, however, we take physiology for our guide in such researches, we shall find support from that unity of character to which every scientific inquiry, and every successive experiment should be reduced.

Pathological tumors afford a good illustration of the extent to which the success of a chemical investigation, and of the method of analysis, depends on a correct physiological view of the question. When we consider the most recent investigations made in relation to this subject, we are led to regard malignant tumors, not as secondary products or parasitic organs, but as exudations which have been arrested in different stages of development and organization. If we adhere to this point of view, we shall no longer attempt to discover the special matters of scirrhus, encephaloid, &c., but shall rather look upon these objects as the means of furnishing us with a clue to the physiologico-chemical processes by which the plasma is developed into cells and fibres, which have hitherto presented insuperable obstacles to the advance of chemical inquiry.

In adverting to the false position assumed by pathological chemistry in reference to pathological anatomy, it must not be forgotten that the pathologico-anatomical school is equally deserving of censure. Whence comes it, we may ask, that those who would set aside pathological

anatomy, and who profess to limit their investigations to the actual facts of medicine, should threaten us with all the horrors of a transcendental humoral pathology? The solution of this question is to be found in the circumstance that, strictly speaking, pathological anatomy is occupied only with the external palpable alterations experienced by the tissues and juices from the action of disease, and that if any of the more gradual stages of transition be made apparent in the course of such processes, these are mere forms or facts, and afford no insight into the *modus* of the organic changes. In a word, pathological anatomy is a purely descriptive science, a natural history of morbid actions, which may lead to the establishment of a system, but not to that of a general principle and to conclusive deductions. It is the geognosy of the morbid organism, and must be allied to a geology of disease, which, however, it is incapable of establishing. It is precisely the purely descriptive character of pathological chemistry that places it beyond the sphere of experiment. Like geognosy, it can only attain its aim—the scientific recognition of objects—with the co-operation of physics and chemistry. If, however, pathological anatomy is to be regarded as the surest foundation of medical science, we must endeavor, on speculative grounds, to ally it more closely with pathology, and thus render it, to a certain extent, more acceptable to the medical public. We are convinced that the principal object had in view by the founder of German pathological anatomy, Rokitansky, in writing the first volume of his celebrated work, which has been so severely criticised, was simply to indicate to pathologists the points of view from which the fruits yielded by the pathological anatomy he had himself established might be most fully comprehended. But it has unfortunately happened that his followers have frequently borrowed from physics and chemistry phrases and modes of representation, without seizing the spirit of these sciences, or even comprehending their methods of operation. Hence there has emanated from this school, notwithstanding the positive observations on which it is based, a multitude of the most unsubstantial medical fictions which, for shallowness, yield to none of the earlier schools. Pathological views in reference to the nervous system (*Nervenpathologie*) have been elevated to the prejudice of physical views (*Nervenphysik*); for here, in consequence of ordinary anatomy being inadequate to explain pathological changes, ideas, or rather mere words, have been unscrupulously borrowed from organic chemistry (by those who were perfectly ignorant of this science) to explain the most complicated processes, of which scarcely anything was known but the final results. Some adherents of the pathologico-anatomical school have presented us with a theory of the crases of the blood in different diseases, although this is a view in which no chemist could at present seriously concur. This theory of crases has been so thoroughly investigated by physiologists in recent times, and its want of foundation made so evident, that we need advert no further to it than to observe that where admixtures and separations are concerned, the chemist is the only competent guide.

A third circumstance which has led to misconceptions in physiological chemistry depends upon an over-estimate of the value of chemical auxiliaries, and a complete ignorance of the present condition of organic

chemistry. Have the numerous analyses of morbid blood instituted during the last few years fulfilled the expectations of physicians? With all due gratitude to the indefatigable investigators who, with no other aid than that which zoo-chemistry could offer, boldly attempted to throw light on those obscure inquiries, it must be admitted that, when we seriously inquire into the recompense of all their labors and sacrifices, we find that the result, although too dearly bought, was altogether inadequate to satisfy the requirements of pathology. Have the numerous analyses of the urine led to much more than the assumption of several new species of disease, or so-called diatheses? Although we might have anticipated greater results, we can hardly wonder that the efforts hitherto made should either wholly or partially have deceived our expectations; for although these investigations may have rendered chemistry no unworthy auxiliary to a physical diagnosis, analyses of morbid products could hardly afford an insight into the chemical laboratory of the organism, while the means were wanting to prosecute them with the scientific accuracy attainable in the case of mineral analyses. Animal chemistry is still wholly unable to afford us a precise, and at the same time a practically useful method of investigating the blood; and how should it be otherwise while we continue to be in doubt regarding the chemical nature of its ordinary constituents? The mineral substances of normal blood are not yet determined, or, at all events, continue to be made the subject of dispute; we scarcely know the names of the fatty matters it contains; one of its most important constituents, fibrin, cannot be chemically exhibited in a pure state; we are ignorant of the nature and mode of secretion of the globulin of the blood-corpuscles; we are still far from being able to separate and determine the so-called protein oxides; and we are also ignorant of the excrementitious substances occurring in the blood. How then, amidst these and a thousand other uncertainties and doubts, can an investigation of the blood be scientifically and trustworthily conducted? We analyze healthy and morbid milk, and yet we are ignorant of the substances whose admixture we have termed casein. The urine, in its morbid condition, presents many varieties; and yet our knowledge of this secretion, frequently as it has been analyzed, amounts to little more than an acquaintance with the quantitative relations of some of its principal constituents; creatinine and hippuric acid have not been determined by any analysis, and doubts are still entertained by some chemists (although most unjustly), regarding the presence of the latter in human urine, while absolutely nothing is known regarding the most important pigment of this secretion. Many experiments have been made and theories broached on nutrition and digestion, and yet to almost the present day the existence of lactic acid in the gastric juice has been contested. Although hypotheses are not wanting regarding the mode of action of pepsin, we know nothing of its chemical nature, and we are wholly ignorant of the proximate metamorphosis of albuminous bodies in the stomach during the process of digestion. Will Mulder be able even with his most accurate analyses, to support his protein theory by the aid of sulphamide and phosphamide? or is this term destined merely to indicate a past epoch of organic chemistry? When such is the state of animal chemistry, can we wonder



that there should be obscurity regarding the chemical processes in the animal body, their various isolated and combined actions, their causal connection and their dependence on external influences and internal conditions? Unfortunately, we might be led to believe, from the lectures and writings of many physicians, that, trusting to the aphoristic and often highly apodictic assertions of certain chemists, they felt secure of having reached the object of their inquiries. Although at present little more than the direction is indicated, we may hope in due time, and after innumerable efforts, to see our endeavors crowned with success.

After having become acquainted with the deficiencies and errors belonging to the chemistry of the vital processes, which was so prominently brought forward at an earlier era, we will now pass to the methods and principles by which alone this science can be made to fulfil its just requirements. The final result of all physiologico-chemical investigations is avowedly that of gaining an accurate knowledge of the progress and causal connection of the chemical phenomena attending the vital processes. To attain to this knowledge, it is not sufficient to detach separate parts from the mechanism of the whole, and to form an opinion of the combined action of so complicated a chemical structure from a more or less superficial examination. Attempts have already been made to establish a splendid theory of the metamorphosis of tissues, but notwithstanding the many able heads and hands that have been engaged in the labor, it is still deficient in the essential of a solid foundation.

It is unnecessary to prove that we must thoroughly understand the substrata of the metamorphosis of the animal tissues before we can venture an opinion on the nature of the processes. The surest supports of physiological chemistry are to be sought, therefore, in general organic chemistry; while the study of the organic substrata of the animal body, or zoo-chemistry considered in the strict sense of the word, must necessarily constitute an integral part of physiological chemistry, and prove a most efficient aid towards its development. If zoo-chemistry ever fulfil its object, it must be by the joint aid of chemistry and physiology; that is to say, individual substances must not only be fully examined in reference to their chemical value and their place in the domain of pure organic chemistry, but they must also be observed in the more general relations which each may bear to the animal organism and its metamorphosis. In a word, the physiological value of each substance should be as carefully considered in zoo-chemistry (the basis of physiological chemistry) as in pure chemistry. It seems to us, that in treating of zoo-chemistry (in the first volume of this work), we shall the best attain this aim by adopting the following arrangement:—namely, by treating of the *chemical* relations of each body, in reference to its properties, composition, combinations, and mode of decomposition, its preparation, the method of testing for it, and its quantitative determination; in explaining the *physiological* relations of each substance, we shall endeavor to determine its occurrence in the animal body, and its origin (whether it be produced within or without the body), and from the above considerations, we shall finally attempt to deduce its physiological value.

We shall treat of the properties of each organic substratum before considering the remaining chemical relations, as it appears to us both

unpractical and illogical to begin with the mode of preparation, as is usually done; unpractical, because no student can comprehend the mode of preparation when he is not in some degree acquainted with the properties of the substance in question, and illogical, because we must have some idea of a body before we can attempt to prepare or exhibit it. The composition of a body must necessarily constitute the most important subject of consideration after its properties and its principal reactions have been duly noticed, for it is only by such means that we can attain to an idea of the nature of a substance, and of the place it occupies in the system of organic chemistry. Hence this section must not be limited to a mere enumeration of analyses or of empirical formulæ, but must embrace a consideration of the arguments that are adducible in favor of the different views of the theoretical internal constitution of a substance, and which are briefly expressed by the rational formulæ. This method is of the greatest importance for the recognition of the physiological relations of organic substances; since without it, we are unable to arrive at any logically correct judgment regarding the origin and the physiological importance of different substances. If a knowledge of the composition of an organic substance were not necessary to the investigation of its *combinations* and *products of decomposition*, we should have placed it after the latter, since they constitute the safest grounds from which we may form an opinion of the rational composition of a body. A careful study of the products of decomposition is however the more necessary, since it is mainly on these that we must base our view of the metamorphoses experienced by any given substance within the vital sphere.

It is only when all these relations have been considered, that we shall deem it expedient to enter upon the different methods of preparation or exhibition, for then only can the directions given for the separation of substances be understood.

Before considering a substance from a physiological point of view, we must examine the means by which we are best able to demonstrate its presence in the animal juices and tissues. The qualitative analysis of organic bodies is still far behind that of inorganic bodies, but attention to this point is the more necessary, since deficient investigations too often lead to hasty and erroneous opinions. Nor does less importance attach to a correct estimate of the methods that have been employed for the quantitative determination of the main constituents of animal fluids; for it is only by this means that we can form an opinion of the value of many of the existing quantitative analyses of physiological and pathological products, and of the conclusions which we are justified in deducing from them.

The physiological consideration of every substance must of necessity be primarily based on its mode of occurrence, for we cannot form any opinion of the importance of a body in reference to the changes of animal matter without knowing where, in what relations, and in what quantity it occurs. When, however, we have examined the origin and decomposition of a substance, we have obtained the firmest base for the explanation of the vital chemical processes.

After having, in this manner, familiarized ourselves with the organic substrata of the animal body, we are still only on the threshold of the

study of the constitution and functions of the animal juices and tissues. Before, therefore, we proceed to the actual study of physiological chemistry (namely, the theory of the metamorphosis of matter, or of the zoo-chemical processes), we take into consideration the substances with which we have already become acquainted in zoo-chemistry, regarding them topographically, in reference to their simultaneous occurrence, and their blending and admixture under the form of animal juices, tissues, and organs. We may extend this classification to the animal fluids as well as to the tissues and entire organs. No one will deny that the knowledge of the chemical constitution of these more complex and frequently variable parts of the animal body is another basis of physiological chemistry, for it is evident that if we would treat of chemical processes, we ought to have a knowledge of the substances implicated in them. This, however, cannot yet be attained in zoo-chemistry in the sense that we attach to this science. We here enter the domain of physiology, in as far as we submit the direct results of physiological actions to an investigation, which however must still be of a purely chemical and essentially analytical character.

The province of chemistry in the consideration of the animal fluids and tissues, is similar to that of mineralogical chemistry, for as in the one case, we seek for elucidation respecting the proximate constituents of often highly complicated compound minerals and rocks, so in the other we endeavor analytically to determine the constitution of animal fluids and solid organized parts by the aid of the knowledge we have already obtained from zoo-chemistry. It was in these data that the nature of physiological and pathological chemistry was formerly studied, and it was believed that the processes themselves might be determined directly from the knowledge afforded by such analyses. The fallacy of such a view is proved no less by the state of our knowledge, some ten years since, regarding the physiology of nutrition and secretion, than by the numerous errors propagated since that period in reference to the chemical processes in the animal body. What were analyses of the blood, urine, milk, and bile before this epoch, but mere isolated facts deficient in those links that ought to bind them to the theory of nutrition and secretion? Physiology then regarded such analyses more as mere accessories than as necessary means for the comprehension of each process. A more exact, although by no means a perfect knowledge of the chemical qualities of these juices was subsequently acquired, and hence it was attempted to establish a more intimate relation between the chemical constitution and the physiological function; but from the absence of a proper analytical foundation, this method not unfrequently led to numerous perversions and dangerous errors, as we have already stated, and as we might illustrate by a large number of examples. Although the results of the chemical analysis of the animal juices may afford many indications of the processes, they by no means enable us to judge of the function itself, however numerous and complete they may be; and it is only by means of experiments founded on the composition of these fluids that we are able to arrive at any satisfactory conclusion regarding the nature of the processes in question.

The study of the zoo-chemical processes based on zoo-chemistry and

the theory of the animal juices, appertains to the third section of physiological chemistry, the theory of the metamorphosis of tissues—of nutrition and secretion. It has already been observed, that the actual object of physiological chemistry is to examine the course of the chemical phenomena of the animal organism in their causal connection, and to deduce them from known physical and chemical laws; or in other words, to explain them scientifically. Even if we regard the chemical substratum, as made known to us by zoo-chemistry and the theory of the juices, in the light of a satisfactorily investigated question, there are still several directions to be pursued before we can reach the proper object of our inquiries. It is here most essential that we should be well acquainted with the paths to be followed, for in our search after truth we are compelled to call to our aid hypotheses which might easily lead us into the domain of pure fiction.

As long as zoo-chemistry and the theory of the juices continue to occupy their present subordinate position, the only method by which the foundation necessary to an exact investigation can be obtained, is that which we may term the statistical. Liebig, Boussingault, and Valentin have indeed, with a more correct view of what was required, attempted to compare the final effects of the whole with the material substrata supplied to the organism. We cannot, it is true, arrive at any conclusion regarding the working of the process itself by a mere juxtaposition and quantitative comparison of the ingesta and excreta of the animal organism, any more than we can judge of the causes and course of diseases by the number of fatal cases recorded: but such experiments furnish us with certain general results which serve as guides to further investigations. Some of the most important questions, whose solution was specially necessary, were unanswerable by any other method. Thus, for instance, it was ascertained, by an accurate investigation of the food, and its comparison with the constituents of the excreta and of the nutrient fluids, that in the ordinary food of animals, albuminous substances occur in sufficient quantity to compensate for the nitrogenous matters lost in the process of nutrition and in the metamorphosis of tissue; while it was thus at the same time shown, that the animal organism does not necessarily possess the property of generating albuminous matter from other substances containing nitrogen. The question whether the animal organism possessed the property of generating fat was answered in a similar manner; and it is well known that by means of such statistical observations (comparing the fat contained in the food with that secreted in the cellular tissue and mixed with the excrements) the contest carried on between Liebig on the one side, and Dumas and Boussingault on the other, regarding the formation of fat, was finally decided in favor of the former.

This statistical method preserves us from setting up untenable hypotheses, and prosecuting useless experiments. How long were the minds of natural philosophers haunted with the illusion that animal bodies possessed the power of generating mineral elements, as lime, iron, sulphur, &c., from other elements, or even from nothing! It was this method alone which exposed the perfect nullity of the obstinately defended dogma of the "vital force."

Statistico-chemical investigations may serve as checks to, or confirmations of other inquiries and methods of inquiry; thus, for instance, Boussingault, by a comparison of the amount of nitrogen in the excrements with that in the food, has fully confirmed the experiments made by Dulong, Valentin, Marchand, and others, which appeared to show that the animal body lost a slight quantity of nitrogen by exhalation from the lungs.

The statistical method would, therefore, appear to be one of the most important aids towards a solution of some of the more general questions in reference to the metamorphosis of the animal tissues. We must, however, be careful not to deduce more from such experiments than what is permitted by the simplest induction; for the results derived from this method have unfortunately too often been made to yield support to the vaguest fictions and the boldest speculations.

It need scarcely be observed that science should not rest satisfied with a knowledge of the final results of chemical processes in the animal body, or with the assertion of the chemical dignity of the vital process *in summâ*, but should be made to enter more deeply into the course of the separate processes, and into the causal relations of phenomena. Here the statistical method cannot of course afford any satisfactory solution to our inquiries; for when we have ascertained by this experimental method that fat is formed in the animal body, we must learn from other methods the manner in which this substance is formed.

The method by which we may examine the course of phenomena and the cause of their succession, might be named *comparative analytical* or *chemico-experimental*, in as far as the chemical phenomena of the living body may be artificially imitated, and the chemical metamorphoses of certain substances external to the vital sphere be compared with those within the influence of the vital processes. Liebig and his school have here done essential service. He was led to believe from his statistical inquiries on fats, that these substances in their transmission through the organism, were in a great measure oxidized and reduced to water and carbonic acid, by which means they specially contributed towards the maintenance of animal heat. As Liebig was by no means inclined to believe, as some have supposed, that fat was consumed in the lungs, somewhat in the same manner as oil burns in a lamp, it was necessary more accurately to investigate its gradual metamorphosis, and its transition through different stages of oxidation, and into bodies containing a larger quantity of oxygen. He believed that he could most readily attain this object by the comparative analytical method; and hence he and his school entered upon a series of experiments on the numerous products of decomposition of fatty matters, and more especially on their products of oxidation; and although we may still be far removed from the object in view, these inquiries have enriched us with many valuable results. A similar instance is afforded by the gelatigenous tissues of the animal body; for although our histological and statistico-chemical investigations leave not the slightest doubt that the gelatin is formed from the albuminous matters, the process of this metamorphosis is still wholly unexplained; and before we shall be justified in forming an opinion regarding this metamorphosis, and expressing it by a chemical

equation, it is indispensably necessary that we should investigate the metamorphoses experienced by albuminous bodies during their gradual oxidation. We are indebted for these views to the admirable investigations prosecuted under Liebig's direction, by Schlieper and Guckelberger, on the products of oxidation of albuminous bodies and of gelatin.

As we learn more thoroughly to investigate the processes of putrefaction and decomposition, and that of the dry distillation of individual animal substances, and therefore the better to understand their regressive metamorphoses, we may hope by this knowledge to arrive at a deduction, based on some probability, regarding their progressive metamorphoses. Among these probable deductions we may place Dessaigne's discovery of the decomposition of hippuric acid into glycine and benzoic acid, Liebig's investigation of creatin, and his pupil's analyses of glycine (glycocoll), which although they do not yet afford us any perfect elucidation of the metamorphoses of animal matter, nevertheless yield many sure points of support for future inquiries on the vital processes.

A third method, which although frequently employed, has hitherto, from the imperfect state of our knowledge, yielded few reliable results, is the *physiologico-experimental*. By this term we would designate that class of inquiries, in which observations are made in the living organism on the result of certain conditions on the progress of a physiologico-chemical process, and on the different stages of that process. We are aware that we shall never succeed in artificially reproducing all the processes as they occur in the living body, since we are here as little able to call forth the necessary conditions and relations, as in the formation of minerals and rocks. It is, therefore, the more necessary to observe a process, of which we cannot judge by imitation, in its course in the living body, and for this end we must chiefly employ natural physiological means. Among these we may reckon the investigations that have been made in reference to the contents of the stomach during the process of natural digestion, to the chemical change of individual substances in the development of the egg during incubation, and to the dependence of the products of respiration on different external conditions. We may further add those experiments that have been made on the changes of individual substances during their passage through the animal organism, or on the effect of different kinds of food, and the metamorphoses of certain nutrient substances during the process of nutrition. To the same method belong all pathologico-chemical experiments, as for instance, observations on the contents of the intestine after the closure of the common bile duct, and on the blood and other fluids after extirpating or tying the vessels of the kidneys. Chemistry, unfortunately, too often fails us to permit of our deriving from this method all the results which it appears to promise; it must, however, ultimately furnish the keystone to all physiologico-chemical inquiries, which, without its aid, would continue insoluble enigmas, and would admit of hypothetical rather than actual explanation. The theory of the metamorphosis of animal matter, without the support of such a physiologico-experimental foundation, must continue to be attended by no little risk.

In conclusion, we would advance a few remarks on the place which physiological chemistry occupies, or at some future period will occupy,

among the auxiliary medical sciences. If the final result of all physio-logico-chemical inquiries be that of comprehending the chemical phenomena of animal life in their different phases and in their causal connections, it is obvious that we must look to this science for a solution of the most important questions of physiology, and of medicine generally. It cannot be denied that most of the phenomena of animal life either consist in or are accompanied by chemical processes; nor can we form an adequate conception of the functions of the nervous system by which sensuous perception and motion are regulated, without the simultaneous existence of chemical actions. For although we are as yet unable to make nervous action fully harmonize with definite physiological laws, or to identify it with certain physical forces or imponderable fluids, all physiological experiments indicate that it is always followed by a chemical reaction, and that the nervous system experiences chemical changes by and through its own activity. It must, indeed, be admitted that any actual proof of such chemical metamorphoses is at present perfectly unattainable, and that our chemical methods would here afford us no higher aid than that which the scalpel yields to the pathological anatomist. But ought we to despair of attaining our object, because we do not as yet clearly perceive the direction we are to follow? Weariness of the senses is the diminished impressibility of the nerves of sense, but its cause cannot reasonably be sought for in any other than a chemical change, experienced by the conducting substance of the nerves. Such a chemical metamorphosis of the nerves of sense from external impressions can no longer greatly excite our astonishment, since we have witnessed the unexpected phenomenon of a picture produced suddenly, and as it were by magic, from the chemical changes effected by the rays of light on an iodized silver plate. Should we not be equally justified in saying that the iodized plate, which after being exposed for a few seconds to a strong light gives only faint and half-effaced images, is wearied like the retina, when after repeated and continuous perception of an image, it gives back only the faint outlines of the object? We may rest assured that the nervous system is not exempt from chemical action; and if the nervous system itself must fall within the domain of chemical contemplation, and a chemical expression remains to be found for its action, no less than for that of digestion and for the formation of blood, it is scarcely necessary to offer further proof of the fact that chemistry is destined to play the most important part in physiology and medicine. However much we may endeavor to exclude chemistry from certain physiological investigations, we shall always find that it involuntarily forces itself upon our notice; for without it we shall be unable to find a physiological equation or a philosophical expression for a process. In a scientific point of view chemistry must, therefore, be regarded as an invaluable acquisition to physiology. We have, then, little cause to dread that Cicero's observation "*Suo quisque studio delectatus alterum contemnit*," will be applied to ourselves, when we assert that physiological chemistry is the crowning point of every physiological inquiry.

When we turn to practical physiology, to pathology, and therapeutics, we are again reminded that chemistry is indispensable. Is there a single disease that is not attended by chemical changes? Can we ever hope to

comprehend or explain the nature of any process, if we are ignorant of its integral factors? Life cannot exist without chemical movements, disease cannot exist without chemical changes. Thus much in reference to pathology; while in respect to therapeutics, it is almost superfluous to observe that chemistry here also plays the principal part, for where has modern pharmacology sought its chief support, save in chemical processes and principles? And if we have advanced so far towards a clear insight as no longer to ascribe supernatural forces to medicines, but to derive their efficiency specially from chemical properties, then must chemistry be the supporting basis of pharmacology. The physician acts upon the body mostly by the aid of matter, which retains its characteristic powers within no less than without the organism. If then nervous action likewise falls within the sphere of chemical metamorphoses, the *Nervina* (or *Neurotica*) of pharmacologists must primarily at least act chemically on this system.

To those who stand on the grounds of exact investigation, holding fast to the fundamental principle that it is from physical laws alone we must deduce a true explanation, and that by induction only can we investigate the causal connection of vital phenomena, no further proof need be adduced of the truth of our assertion that physiological chemistry occupies the highest place among the sciences auxiliary to medicine. Even those who deem special forces and special laws necessary to the explanation of vital phenomena must admit that chemical methods are the most important for the investigation of these actions, and for the solution of such questions, if, as indeed cannot be denied, it is only by a thorough investigation of the physical forces acting in the living body that we can become acquainted with a true vital force or vital law. With those who judge of vital forces by subjective feelings, and would stamp nature with the impress of their own ideas, we will not contest the point of view we have adopted; but leave them to regard chemistry, like physics and anatomy, as a mere auxiliary towards an adequate appreciation and contemplation of nature.

It now only remains for us to add a few words on the relation of pathological to physiological chemistry. Neither from a theoretical nor a practical point of view can we concur in the assertion that pathological chemistry is separate and different from physiological chemistry. Experience shows us the impracticability of such a separation, for how much mental energy has been wasted, as it were, in the investigation of unattainable things; and among these we may class pathological chemistry, when not based on physiological principles. It would assuredly be going too far, to assert that the natural inquirer should undertake no experiment that could not afford a definite solution to a well-grounded question; but it must be admitted that there is an almost countless number of pathologico-chemical experiments which have yielded no result, and which obviously could yield none; and indeed it seems scarcely comprehensible that we should attempt to understand that which is abnormal, while we continue ignorant of that which is normal. Before we can institute a comparison between two things, we must be familiarly acquainted with at least one. Here we do not by any means wish to maintain that no pathologico-chemical inquiries should be prosecuted,



for this would be as absurd as to withhold our attention from pathology until we supposed ourselves fully enlightened on the subject of physiology. We would, on the contrary, limit our objections to those analyses of pathological products which have no relation to any one leading idea, are devoid of connection with any scientifically established fact, and do not bear upon general chemical or physiological propositions. Such investigations are so numerous, that our weekly periodicals are seldom without one or more analyses of diabetic urine. These results would, doubtless, afford additional proof of the well-established fact that sugar is present in diabetic urine, if we did not feel assured that the diabetes was not diagnosed until the existence of sugar had been demonstrated in the urine. We seldom meet with any observation on the relation existing between the quantity of sugar excreted in a given time, and the quantity of food taken during the same period; while other and similar considerations of equal importance are also usually disregarded.

The severance of pathological from physiological chemistry is even less admissible in a scientific than in a practical point of view. We will not here pass judgment on the obscure abstract idea of disease, but whatever value such a view may have in reference to life and medical practice, and however pathologists may strive artistically to define it, it must continue illogical in reference to theory and science. But whatever view we may here adopt, it must be admitted that pathological and physiological chemistry cannot exist independently,—a view requiring no circumstantial proof. The power and the law remain the same, whether the points of application be more or less remote from the fulcrum of the lever; the result alone is different. Pathologico-chemical phenomena do not originate in the occurrence of new forces or special laws, but merely from the chemical points of application being somewhat different; that is to say, the relations are changed under which the substrata develop their actions of affinity. Pathological phenomena can, therefore, only be recognized when manifested preponderatingly in some one direction, but they of necessity obey one and the same law. As the result of indispensable conditions we cannot then regard them as anomalous or abnormal. If protoxide of iron is no longer precipitable by alkalis when organic acids are present, and if fibrin loses its capacity for coagulating in the presence of certain salts, we no more apply the term *diseased* to these substances than to a clock which stops because the weight has run down. When, in consequence of any influence, the capillaries become dilated, and the blood contained in them stagnates, exudes, or coagulates, we do indeed recognize the occurrence of something singular and not of ordinary occurrence, but nothing independent of a law. The physician may designate inflammatory symptoms as abnormal and morbid, but the philosophical inquirer sees only the necessary result of laws acting under different relations, for he has to deal only with fixed laws and not with rules abounding in exceptions. The chemist is an investigator of nature even when occupied in studying pathological processes, as the physiologist is still engaged in physiology, when turning his attention to the less frequent phenomena of the living body, for there is no special science for the exceptional phenomena of nature but only one physiology as there is one all-powerful law of nature.

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We are tempted, notwithstanding the above observations, to cast a glance at the position occupied by physiological chemistry, in relation to what is called metaphysiology. The recent advances of organic chemistry have unfortunately been interwoven with a fantastic physiology, which designates itself as a comparative science. This is not a science comparing together the functions of the organs of different animals, as comparative anatomy compares their structure, but a system founded on abstractions and ideal comparisons; that is to say, on figurative representations of subjective conceptions, in which the results of objective investigation are advanced in defiance of the most contradictory facts. We entertain all due respect for that form of metaphysics which occupies the same rank among the speculative sciences as physiology and chemistry hold among the exact sciences. Metaphysics and physiology resemble two diverging lines, which coincide only in their starting-point, and differ so widely at all other points, that they cannot be united unless to the detriment of true science. The physicist has maintained his stand more firmly and securely than the speculative natural philosopher, who never relaxed in his attempts to force his complex ideas, constructed according to a subjective standard, upon the objective experiments of the physicist. On this principle it has been attempted to anticipate intellectually the discoveries and general propositions which the physicist endeavors to attain by practical evidence, and thus science has been confused in a manner that cannot fail to retard its advance. There are now indeed but few remaining followers of the school of speculative natural philosophy, which emanated from the same exaggerated bias of the age which in poetry gave rise to the romantic school. Men created for themselves an Ideal, to which they gave the name of nature.

Although such a system of metaphysics<sup>1</sup> completely mistakes its province, it is yet essential that "the chemist should raise himself above the vital, no less than the chemical process, in order to compare them both in their principal properties and results, and to represent them in their co-existence, founded as it is in objective processes." This is, however, a point of view from which no mere chemist should observe the phenomena of nature; for no exact investigation is compatible with imaginative speculation, which can exhibit only artificial comparisons and obscure reflections of dimly comprehended physical phenomena. We have not hesitated to avow that we have assumed a thoroughly radical point of view, in reference to specific vital phenomena and vital forces; for we cannot rest satisfied with the mysterious obscurity in which they have been artificially enveloped. With the physicist we would uphold the reality of phenomena; and while we admit that the consciousness of the reality of matter is only the result of an abstraction, we must regard this abstraction, by which we recognize the Immaterial, the Spiritual, and the Force, as originating in reality. We therefore believe, with the diffidence befitting a genuine student of nature, that it would be wiser and more conducive to the spread of true knowledge to adhere, in the study of vital processes, to matter, and to the laws by which it is determined, than (following the fictitious abstractions of dynamical processes)

<sup>1</sup> Geubel, *Grundzüge der wissenschaftlichen Chemie*, Frankf. a. M. 1846, and L. Müller, *Berzelius's Ansichten*, Breslau, 1846.

to assume that there exists in life a higher power of the spiritual force pervading matter. While, therefore, in opposition to the views of these natural philosophers, we must refer all force to matter, we have no fear of degrading "vital phenomena to mere mechanical, physical, and chemical processes;" since our most exalted conception of nature and the sublimest natural philosophy emanate from the very simplicity of physical laws, and the unlimited variety of phenomena to which they give rise.

We are firmly convinced, that even metaphysiology will be unable to deprive physiological chemistry of the consideration due to it among physical studies, in its explanation of vital processes; and we will therefore leave it to the poetic and the imaginative to depict the romance of the protecting activity and sturdy contest maintained by the vital force, and of a struggle between different powers,—between the attraction and repulsion of polarities. Does it not need a superabundant richness of fancy to believe, with metaphysiologists, that apparent death, trance, or (as it has been termed) latent life, is the predominance of the spiritual over the material (the metamorphosis of matter being at its minimum), rather than a predominance of the material over the spiritual, as sounder minds would be led to assume? It would be well if these spiritualists would look down from the high stand they have chosen, and deign to believe that there are some among those experimentalists, who, clinging to matter, and gathering their facts with ant-like industry from the lowly earth, notwithstanding that they have long held communion with the poet-philosopher, Plato, and the philosophical natural inquirer, Aristotle, and have some familiarity with the Paraphrases of Hegel and Schelling, are yet unwilling to relinquish their less elevated position. If these happy admirers of their own Ideal had descended from their airy heights, and closely examined organic and inorganic matter, they would not have deemed it necessary to assume, that besides carbon, hydrogen, nitrogen, and oxygen, organic substances must also contain an *organogenium*, or latent vital force, or whatever else they may be pleased to call it. Had they sought information from a chemist, they would have learnt, that when exposed to the clear light of rigid logic there is no essential difference between organic and inorganic bodies: a chemist, totally unacquainted with organic matter, would *a priori* have deduced all these incidental differences of matter from the doctrine of affinity and the science of stoichiometry, evolved from dead matter. However these advocates of a romantic poetry of nature may despise the swarm of industrious investigators, who are often unwearingly occupied for years together in endeavoring to collect a few firm supports for the great edifice of a true philosophy of nature, we do not despair of seeing our work rise in simple grandeur, more durable and lasting than those sophisms of natural philosophy, which, passing through ages, from Pythagoras and Empedocles to Schelling and Hegel, have, like the sand of the ocean shore, been alternately upborne by one wave and engulfed by the next.<sup>1</sup>

<sup>1</sup> If any of my readers have chanced to meet with the article "Chemismus in der Medicin," which appeared in the "Gegenwart," they have probably been struck by the similarity existing between the ideas expressed in the present work and the line of thought followed in that essay; I therefore feel called upon to avow the authorship of it.

## THE

## ORGANIC SUBSTRATA OF THE ANIMAL ORGANISM.

WHILE we admit that the general investigation of nature must derive its chief support and stability from the investigation of particulars; and while we deplore the evils that have accrued to the natural sciences from the premature abstractions and hazardous generalizations deduced from data, which are in themselves correct; we must remember, that no department of natural science, however limited its domain, should be entered upon without the aid of certain leading maxims, and without a definite aim. These must be sought by physiological chemistry in physiology, no less than in general chemistry; for without these aids zoo-chemistry will continue a confused mass of loosely connected facts, from which every fanciful inquirer may select whatever suits his views, to beguile himself or others with short-lived dreams or illusions.

The general principles and recent acquisitions of chemistry are as essential to the consideration of the properties and chemical metamorphoses of animal substances, as an intimate acquaintance with physiological theories is to the deeper insight into the chemistry of the animal functions. It would be both inappropriate and detrimental to this branch of science to borrow from general chemistry only such matters and facts as refer to the animal body, in order to accumulate a mass of disjointed bodies, and group them together simply according to their physiological import; as if we considered zoo-chemical processes in a purely chemical light, depending upon combination or decomposition, on chemical dualism, the theory of acids and bases, &c.: we should rather adhere in our study of the chemical substrata of the animal organism to the more general chemical points of view, from which we may consider the chemical nature of these heterogeneous substances; or, in fine, we must not leave it to chance in zoo-chemistry whether or not we examine a chemical substance according to its occurrence in, or absence from the animal organism. We must pay special attention to the place occupied by each member of the group of chemical substances; while the contiguous members and allied substances, that may not have occurred in the same order in other animal bodies, must not be disregarded. It would be illogical to regard the metamorphic products of those animal matters that we have not hitherto been able to detect in the excreta of animal bodies, as excluded from zoo-chemistry; or, at all events, as constituting only a less essential and more supplementary portion of the science. Zoo-chemistry

should not only embrace, according to the principles of pure chemistry, all substances standing in a more or less intimate relation to the matters actually found in animal bodies, but it should likewise make the fullest and most extended application of the various propositions and theories by which general chemistry has at different times been enriched. At the first glance it might appear as if the physiological momentum were entirely lost in such a conception of zoo-chemistry; but so far from this being the case, we find that by such a method physiology is made to afford the greatest aid.

*The physiological importance of a body is mainly dependent on its chemical composition and quality.* If this proposition be true, the assertion that a chemical conception of animal substances must likewise be a physiological one, can no longer be called in question. The physiological capacities of the material substrata of animate beings can be referred only to their chemical qualities, and no form of physiology, that was not tinged with sophisms of the spiritualist school, could hold that a chemical substance should depose all its integral properties in the animate body, to assume higher or more spiritual capacities in the vital sphere. But while we would endeavor in the following pages to establish the principle of the purely chemical arrangement of zoo-chemical substances, we at the same time most fully award to physiology what is its due. A chemical arrangement of animal substances must be in perfect accordance with a physiological one; while the latter would neither be rational, correct, or in accordance with nature, if it were to associate substances having different chemical qualities, and artificially separate others of analogous chemical characters. Thus, it is self-evident, that substances containing no nitrogen, as starch, sugar, &c., must be associated with very different physiological functions from albuminous bodies, containing a large quantity of nitrogen: but we should hardly have expected that the difference between nitrogenous and non-nitrogenous bodies should be so clearly shown in the two great kingdoms of living organisms; the vital phenomena of animals and plants, in a great measure owe their differences to the diversity of these two classes of chemical substances. We shall find in the course of our observations, that pure chemistry cannot sever or group together organic substances, otherwise than as physiological conditions shall require.

When we speak of applying a purely chemical principle to the classification of the objects embraced in zoo-chemistry,—understanding by the term, the theory of the chemical substrata of animal organisms,—we do not refer to the old and bygone classification of organic substances into acids, bases, and indifferent or amphoteric bodies; for we are of opinion that a classification of animal substances, according to their combined chemical relations and their chemical import (but not according to a single property, as for instance their basicity or acidity), must be physiologically correct, since it is a natural method of arrangement. On the other hand we regard a purely physiological principle of classification in zoo-chemistry (such as we followed in the first edition of the present work) as no less irrational and unnatural than that which has originated in views based merely on a theory of affinities.

Although we might at first sight be disposed to regard as appropriate a classification of organic substrata into nutrient matters and excreta, the practical application of such a mode of treatment will exhibit numerous deficiencies, which completely nullify the advantages it might have been supposed to possess. For it soon becomes apparent, that a body which appears in one part of the animal organism, or in one process, strictly as a product of decomposition, is applied in another to the formation of a tissue, or the accomplishment of a purely physiological function. A separation of zoo-chemical substances into secreted and excreted matters, leads to the greatest uncertainty and the most intricate confusion. We must, however, admit that every systematic mode of arrangement seems impracticable in a purely empirical science, which ought only to follow a genetic or ætiological, and not a teleological method; since the latter can, at most, only indicate the direction in which investigation should be pursued in an immature science. A new phrase has, however, been recently employed by which it was conjectured that zoo-chemical processes might, according to their nature, be separated into two wholly different classes, viz. progressive and regressive metamorphosis of matter. However deserving these words may be of being retained in physiological chemistry to serve as concise and generalizing designations, they do not express definite ideas in relation to the abstruser study of this science, or of pure zoo-chemistry. Without dwelling upon the fact that it is impossible to prove, in the case of many zoo-chemical substances, whether they belong to the progressive or the regressive metamorphoses of matter, we will only observe, that even in the animal processes no limits can be drawn between the termination of progressive and the commencement of regressive metamorphosis. Carnivorous animals only introduce into their organism well-elaborated animal matter, and hence in them the extent of the progressive metamorphosis must be very inconsiderable; yet an opinion has long been entertained, that in animal life there is a regressive formation alone, and in vegetable life only a progressive development of organic matter. The acrimonious discussion that arose, as to whether the fibrin of the blood belonged to the progressive or the regressive metamorphosis, is sufficient proof that no leading principle is embodied in these terms. We perceive, therefore, that a purely physiological mode of classification is as untenable as those chemical methods which have been borrowed from the individual, and, in most cases, incidental properties of substances.

No chemist at all acquainted with the present state of organic chemistry, will be disposed to place such bodies as albumen and urea in one genus, because both these substances are nitrogenous and amphoteric, any more than the physiologist, who is well aware that a nutrient substance must of necessity have a very different chemical constitution from an excreted substance. We would, therefore, again observe that chemists and physiologists must perfectly coincide in their views respecting the mode of classifying and considering animal bodies, and that where they differ in their description, both cannot be true to nature; for where, for instance, a physiologist should regard a substance as a product of secretion, while the chemist classed it with albuminous substances in accordance with his observation of its constitution, one or the other must be in

error; since the chemical qualities of a body cannot be at variance with the physiological. That method which fulfils the requirements of both sciences, chemistry as well as physiology, can therefore be the only correct mode of treating zoo-chemistry.

Although zoo-chemistry constitutes the firmest basis of physiological chemistry, and although the chemical element should be duly considered, we ought not wholly to lose sight of the physiological relations of individual substances. It is not enough to describe the properties, composition, preparation, and decomposition of matters without also considering their physiological character. The occurrence of a substance in certain parts of the animal body and in certain processes, its relation to the general metamorphosis of matter, and its progressive or regressive formation, are all questions for whose solution we do not look to pure chemistry, although physiology alone is equally incompetent to the task.

A structure such as we have endeavored to sketch, appears to us indispensable to zoo-chemistry, before we can expect that physiology and medicine will furnish an exact reply to those general questions in chemistry which refer to the more important processes. Similar views have undoubtedly guided most true natural inquirers in their labors in this field of scientific investigation. Nor have such men as Berzelius, Wöhler, Liebig, and Mulder, ever undertaken investigations which from their deficiency in all scientific bases could not lead to any scientifically reliable results. We find that such men have always endeavored to afford that internal scientific support to pure zoo-chemistry without which it must continue a mere medley composed of disjointed facts. In the present day we are, however, justified in expecting well-grounded physiological results from pure zoo-chemistry, nor do we exaggerate in stating that more light has been thrown on the metamorphosis of animal matter by such zoo-chemical investigations, as Mulder's on albuminous substances, Liebig's on creatin, and Wöhler's on uric acid, than by many hundred analyses of the blood and urine.

In accordance with the views already advanced, we shall in the following sketch of the zoo-chemical elements, retain those groups that have been established by the most recent investigations of pure chemistry. Bodies of homologous chemical value must also possess common physiological relations. We shall begin with bodies of the simplest composition, most of which have seldom, if ever, been found developed in the animal organism; but with which it is necessary we should become acquainted as the derivatives of animal substances. By thus passing from the groups of simply constituted bodies to those of more complicated composition, we shall gradually become more familiar with the mechanism of the association and separation of organic matter, until we are finally enabled to form a correct judgment of the most complicated substances of the animal organism. We must, however, submit the facts before us to a careful and critical inquiry, if we would employ zoo-chemistry as the firmest support of physiological chemistry. For there is scarcely any department of scientific inquiry in which truth and error, suppositions and facts, acquired and presumed results, and positive and hypothetical deductions, have been more confounded. We need only refer to the fanciful trifling with chemical formulæ which, from bearing the impress

of the words and symbols of an exact science, have deceived many unaccustomed to such characters. The cause of the many erroneous views which have passed from physiological chemistry to physiology and medicine, mainly depends upon the inadequate knowledge of what is necessary for *the establishment of a formula for the chemical constitution of a body*. It seems, therefore, not wholly inappropriate, in an introduction to zoo-chemistry, to refer to the points in pure chemistry, from which alone the chemist is able to deduce a formula.

We might indeed draw some conclusions regarding the atomic composition of a body from the mere result of one or more elementary analyses, or, in other words, we might, from the percentage composition of a body, construct an empirical formula which would serve to exhibit the relation of the separate elements to one another. But this method can alone possess any scientific value when, on the one hand, we are convinced that the substance under consideration is chemically pure, and when, on the other, after the former fact has been fully proved, the errors incidental to every analysis are considerably smaller (*i. e.* when the variations in the percentage results of the analysis are less) than would be afforded by any other formula than the one calculated. Such variations by which an entire analysis may be rendered unavailable are of common occurrence in the determination of hydrogen; the atomic weight of this element being so small that the slightest variations in the percentage composition derived from the individual analyses may cause the formula of a body to differ by one or more atoms of hydrogen. Moreover, another reason why elementary analyses often exhibit the most marked variations in the quantity of hydrogen, is that the drying of an organic substance is only relative, and as many of these substances are extremely hygroscopic, it is impossible, even with the greatest care, to prevent them from condensing water from the atmosphere during the process of weighing. We call this drying relative, because in many substances we are unable to determine at what degree of temperature, and after what time they should be regarded as dried, as decomposed, or as still retaining water. Hence it is evident that the number of atoms of hydrogen will be computed with the least certainty in the most important elements of zoo-chemistry, as in the albuminous matters and their derivatives, which are bodies of very high atomic weight.

In consequence of the atomic weights of these substances being so high, and considering the great uncertainty whether they are free from all admixtures, excepting the salts with which they are inseparably connected, the number of atoms of carbon cannot be computed with certainty from the empirical result of the analysis. As, moreover, we possess no means of directly determining the oxygen contained in an organic body, and can only estimate it by the loss in the weight of the substance analyzed, that is to say, by the subtraction of the quantities of carbon, hydrogen, and nitrogen, the collective errors in the investigation will frequently affect the number representing the oxygen, which must therefore be regarded as the most uncertain number in the analysis.

When all the errors which attach to the calculation of atomic formulæ

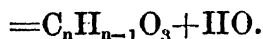


from the direct results of elementary analyses have been as thoroughly as possible avoided, and even when they may be regarded as  $= 0$ , the formula will still only have a *problematic value* until the saturating capacity of the body has been determined by direct experiment, that is to say, until the atomic weight derived from the saturating capacity of the body shall be found to accord with that deduced from the analysis. We have therefore no guarantee for the true atomic weight of a body, or for its atomic composition, without a previous knowledge of the saturating capacity, even supposing that all the other data were perfectly correct, and free from doubt. Thus, for instance, we should not know whether lactic acid and starch were composed according to the formula  $C_6H_5O_5$ , or  $C_{12}H_{10}O_{10}$ , or according to other multiples. But there are, unfortunately, many animal substances of a higher order, whose atomic composition cannot be tested by a comparison with their saturating capacity. Such substances either do not combine in definite proportions with other substances, or do so in various relations, so that it is impossible to determine which combination is actually to be regarded as the neutral one. The variations in the numbers of the saturating capacity, are frequently much more important in such bodies (partly owing to the admixture of mineral substances with them) than those of the numbers of the elementary analysis; that is to say, the atomic weight derived from the saturating capacity is frequently no less uncertain than that derived from the elementary analysis.

If these well-established rules be followed, and the properties of most albuminous matters and their derivatives be compared in accordance with these considerations, we shall easily perceive what credit should be attached to the formulæ established for the composition of these bodies, and with what temerity these most problematic of all formulæ have been transferred to physiology only to involve it in a new labyrinth of vague dreams and fantastic fictions. This absence of reasoning power, this perfect ignorance of all leading maxims having any scientific import, this superficial knowledge of the true requirements of science, has led many physicians to make elementary analyses of admixtures of several substances of a highly variable composition: as, for instance, of blood, bile, muscle, &c., and to establish chemical formulæ from the data thus afforded. Even were it not known that these animal fluids are composed in their physiological condition of constituents having very variable and different proportions, and that microscopic observation had shown the muscular bundles to be composed of very distinct and separate morphological elements, this offence against the first principles of chemistry ought not to be palliated, on the supposition that unchemical experiments might chance to yield valuable physiological results; for physiology demands from chemistry exact and scientifically established facts, and not the mere *ignes fatui* of chemical illusions.

## NON-NITROGENOUS ACIDS.

## THE BUTYRIC ACID GROUP.



The acids of this group possess (as is indicated by the above formula) the following property; in their isolated state, that is to say when not combined with bases, they contain 4 atoms of oxygen and a multiple of a carbo-hydrogen polymeric with olefiant gas; in their combination with bases they lose, however, 1 atom of water, so that the resulting salt contains an acid in which 3 atoms of oxygen are combined with a carbo-hydrogen whose hydrogen is always too little by 1 equiv. exactly to produce olefiant gas with the carbon.

The number of this class of acids is considerable; we have

Formic acid,	.	.	.	.	.	.	$C_2H_3O_3.HO=(CH)_2O_4.$
Acetic acid,	.	.	.	.	.	.	$C_4H_7O_5.HO=(CH)_4O_4.$
Metacetic acid,	.	.	.	.	.	.	$C_6H_9O_5.HO=(CH)_6O_4.$
Butyric acid,	.	.	.	.	.	.	$C_8H_{11}O_5.HO=(CH)_8O_4.$
Valerianic acid,	.	.	.	.	.	.	$C_{10}H_{13}O_5.HO=(CH)_{10}O_4.$
Caproic acid,	.	.	.	.	.	.	$C_{12}H_{15}O_5.HO=(CH)_{12}O_4.$
Enanthic acid,	.	.	.	.	.	.	$C_{14}H_{17}O_5.HO=(CH)_{14}O_4.$
Caprylic acid,	.	.	.	.	.	.	$C_{16}H_{19}O_5.HO=(CH)_{16}O_4.$
Pelargonic acid,	.	.	.	.	.	.	$C_{18}H_{21}O_5.HO=(CH)_{18}O_4.$
Capric acid,	.	.	.	.	.	.	$C_{20}H_{23}O_5.HO=(CH)_{20}O_4.$

Closely approximating to them in their composition is another somewhat extensive group of organic acids, the "fatty acids," which, however, we shall consider separately, because they possess certain distinctive characters which would interfere with the general view which we propose to take of these acids.

It is not surprising that as these acids present a perfect analogy in their composition (homology), they should also present very many similarities in their physical and chemical properties. They are all fluid at an ordinary temperature, and, when freed as much as possible from water, are mostly oleaginous; they do not crystallize and solidify at a higher temperature than  $0^\circ$ , but are so volatile that at an ordinary temperature they more or less powerfully irritate the eyes and nostrils; they are colorless, but have a peculiar burning or acrid taste. They are soluble in almost every proportion in water, alcohol, and ether; they redden litmus powerfully; they may be distilled without being decomposed; their boiling-point ascends with the number of the atoms of the carbo-hydrogen (according to Kopp, at the rate of  $19^\circ$  [ $34.2^\circ$  F.] for 2 atoms of CH), and the densities of the vapors of these acids have a similar relation to the number of the atoms of the carbo-hydrogen; moreover these vapors are inflammable when too much aqueous vapor is not mixed with them.

Combined with bases, these acids form salts which are for the most part soluble, and some of which crystallize readily. With organic haloid

bases,—the oxides of methyl, ethyl, amyl, and lipyl,—they form what are called haloid salts, which are produced either by direct union of the acid and the base, or by double decomposition. Almost all the compounds of the first three are liquid, and extremely volatile; their boiling-point is lower by a definite number of degrees than that of the corresponding acids when deprived as thoroughly as possible of water. In no class of bodies have so large a number of metameric substances been hitherto found as in this; thus, for instance, metacetic acid= $C_6H_5O_3.HO$ , formate of oxide of ethyl= $C_4H_5O.C_2HO_3$ , and acetate of oxide of methyl= $C_2H_3O.C_4H_3O_3$ , containing equal numbers of the atoms of the individual elements= $C_6H_6O_4$ , are metameric; so also are cenanthylic acid= $C_{14}H_{13}O_3.HO$ , acetate of oxide of amyl= $C_{10}H_{11}O.C_4H_3O_3$ , caproate of oxide of methyl= $C_2H_3O.C_{12}H_{11}O_3$ , and valerianate of oxide of ethyl= $C_4H_5O.C_{10}H_9O_3=C_{14}H_{14}O_4$ .

Most of these acids were formerly called *volatile fatty acids* from having first been made known through the decomposition of many fats; but this designation ought no longer to be retained, because while a large number of these acids cannot be prepared from fats, others again may be obtained with equal facility, as educts and products of many other animal or vegetable substances. Thus, for instance, butyric acid, which was formerly regarded as the representative of these acids, may be as easily obtained by the putrefaction or artificial oxidation of albuminous substances, or by the fermentation of sugar and starch, as by the saponification of butter.

Before we enter upon the consideration of the individual acids belonging to this group, we must draw attention to some of the relations possessed in common by all of them, and which depend upon the substances with which they are intimately connected, upon the series of homologous bodies from which they are either produced, or into which they are converted under like conditions, and more especially upon their chemical constitution.

We would first draw attention to the fact that by following the theory of organic radicals, we discover a number of bodies which may be regarded as lower stages of oxidation of the carbo-hydrogen radical of these acids. Thus we have bodies of the general formulæ  $C_nH_{n-1}O + HO[=(CH)_nO_2]$  and  $C_nH_{n-1}O_2 + HO[=(CH)_nO_3]$ . The substances composed in accordance with the first of these formulæ have been named oxides of the radicals of the acids, or more commonly aldehydes. These bodies are for the most part liquid, very volatile, and oxidize rapidly when exposed to the air, becoming thus converted into their corresponding acids. Up to the present time, the following bodies of this class have been accurately studied.

Aldehyde of acetic acid,	.	.	.	.	.	.	$C_2H_3O.HO$ .
Aldehyde of metacetic acid,	.	.	.	.	.	.	$C_4H_5O.HO$ .
Aldehyde of butyric acid,	.	.	.	.	.	.	$C_6H_7O.HO$ .

The stage of oxidation= $C_nH_{n-1}O_2.HO$ , existing between these oxides and the acids in question, is only found in a few cases; as

Acetylous acid,	.	.	.	.	.	.	$C_4H_5O_2.HO$ .
Cenanthylous acid,	.	.	.	.	.	.	$C_{14}H_{13}O_2.HO$ .

Moreover they are rapidly oxidized by the air, and converted into the corresponding acids.

From the dry distillation of the baryta-salts of several of these acids, substances isomeric with the aldehydes have been obtained. They are known by the terminal syllable *al*; they occur as oily, very volatile, pungent fluids, which can be distilled without undergoing decomposition, dissolve freely in alcohol and ether, but not in water, possess neither acid nor basic properties, are not so easily converted into the corresponding acids by the action of the atmosphere as by means of oxidizing substances, and readily exchange a portion of their hydrogen for chlorine. At present we are acquainted with—

Butyral, . . . . .	$C_8H_8O_2$ .
Valeral, . . . . .	$C_{10}H_{10}O_2$ .
Cenanthal, . . . . .	$C_{14}H_{14}O_2$ .

Another series of derivatives is obtained from these acids by heating their salts with strong bases, the acid losing the elements of an atom of carbonic acid, and becoming converted into a substance which, in addition to a carbo-hydrogen polymeric with olefiant gas (but composed of an odd number of atoms), contains 1 atom of oxygen; thus, for instance,  $CaO.C_8H_7O_3 - CO_2 = C_7H_7O$ . These bodies are distinguished by the terminal syllable *one*; they are colorless and very volatile oils with a penetrating odor, readily soluble in alcohol and ether, insoluble in water, very inflammable, and not capable of combining with acids or bases.

In these acids, as in many other organic bodies, certain atoms of hydrogen may be replaced by the corresponding number of atoms of chlorine, bromine, or iodine; thus, for instance, the formation of chloracetic acid is explained by the equation  $C_4H_3O_3.HO + 6Cl = 3HCl + C_4Cl_3O_3.HO$ . In butyric acid, various numbers of atoms of hydrogen may be replaced by an equal number of atoms of chlorine; thus, we have two chlorobutyric acids represented by  $C_8(H_5Cl_2)O_3$ , and  $C_8(H_3Cl_4)O_3$ . However strongly Berzelius, even to the very close of his life, may have contended against the substitution-theory, yet we must not disregard it in the consideration of the constitution of organic bodies. For although this mode of indicating the composition of organic bodies containing chlorine is opposed to the electro-chemical views that have hitherto prevailed in chemistry, it ought not to be wholly rejected, since it is the mode of representing the constitution of such bodies, which approximates most closely to the empirical composition. It necessitates no rigorous adhesion to the metaleptic views of Dumas and Laurent, if for the sake of greater facility of inquiry, and a better comprehension of the subject, we employ this mode of representation, and arrange the formulæ of these bodies so as to substitute chlorine in the place of hydrogen.

But putting out of the question the practical advantages afforded by this mode of viewing the subject, and independently of the circumstance that Berzelius's mode of indicating the composition of such bodies is very far-fetched, and cannot without great difficulty be brought in accord with other experiments, this mode of investigation is recommended by the circumstance that, in most cases, notwithstanding the loss of atoms of hydrogen, and the introduction of negative chlorine, bromine,

or iodine, or of the complex atom= $\text{NO}_4$ , corresponding to hyponitric acid, the new body retains the chemical character of the original compound; that is to say, if the mother-substance were an acid, the newly-formed substance would be so also; if it were neutral, the new compound would likewise be neutral; and it is very remarkable, that basic bodies, like the alkaloids, continue bases when the above elements, or hyponitric acid, are substituted for the atoms of hydrogen.

All the acids of this group likewise form *amide-compounds*. The term *amide* is known in inorganic chemistry. The atomic group  $\text{H}_2\text{N}$ , which cannot be exhibited in an isolated state, is found in many metallic preparations produced by treating compounds of the metallic oxides with ammonia. It might thence be assumed that the atom of oxygen of the metallic oxide, as for instance of the oxide of mercury, has united with an equivalent of hydrogen of the ammonia to form water, and that the metal then unites with what remains of the ammonia= $\text{H}_2\text{N}$  to form the so-called amide. In organic chemistry the amides are produced in a similar manner, with this difference only, that in this department it is chiefly acid substances which have a tendency to enter into such combinations. We can best realize the production and decomposition of organic amides, by assuming that the hypothetical anhydrous ammonia salt of the organic acids loses an equivalent of water, while an equivalent of hydrogen is withdrawn from the ammonia, and an equivalent of oxygen from the acid. Thus acetamide is equal to acetate of ammonia, minus 1 atom of water, since  $\text{H}_3\text{N} \cdot \text{C}_4\text{H}_3\text{O}_3 - \text{HO} = \text{H}_2\text{N} \cdot \text{C}_4\text{H}_3\text{O}_2 = \text{C}_4\text{H}_5\text{NO}_2$ .

According to the theory of substitutions, one atom of the oxygen of the acid in these combinations is replaced by the complex atom  $\text{H}_2\text{N}$ ; but this mode of viewing the subject cannot be adopted, since the acids, by this union, entirely lose their acid character, and even basic bodies, on their entering into combination with amide completely lose their basicity. The knowledge of these amide-compounds, and of their general characters, which have only recently attracted the attention of chemists, is of great importance, because there is reason for believing that several substances occurring in the animal and vegetable kingdoms belong to this class of bodies.

While the amides of many other acids can be artificially produced, by the exposure of the ammonia salt to heat, or by the treatment of the chlorine-compounds with ammonia, the amides of the acids of this group are best obtained from their salts of oxide of ethyl and ammonia. Thus acetamide is formed on digesting acetate of oxide of ethyl (acetic ether) with fluid ammonia, since  $\text{C}_4\text{H}_5\text{O} \cdot \text{C}_4\text{H}_3\text{O}_3 + \text{H}_3\text{N} = \text{C}_4\text{H}_5\text{O} \cdot \text{HO} + \text{H}_2\text{N} \cdot \text{C}_4\text{H}_3\text{O}_2$ .

As is shown in this formula, the oxide of ethyl becomes converted in this process into the hydrated oxide, or, in other words, the ether becomes converted into alcohol; the water necessary for this change is formed from 1 atom of the oxygen of the acetic acid and 1 atom of the hydrogen of the ammonia.

The amides of these acids are solid, crystallizable, and colorless; they are soluble in water and alcohol, sublime without undergoing decomposition, have no action on vegetable colors, and are indifferent towards weak

acids and bases. If, however, they be treated with strong acids or bases, they assimilate water and become decomposed into ammonia and the corresponding acid.

Acetamide, treated with caustic potash, yields ammonia and acetate of potash:  $C_4H_5NO_2 + KO.HO = KO.C_4H_3O_3 + H_3N$ .

The behavior of this amide, as well as that of all others, towards nitrous acid, is very characteristic; for, by the action of this acid, these amides are converted into the original acids, ammonia being at the same time developed. (Piria.<sup>1</sup>)

We may explain this process by supposing that hydrogen is assimilated through the action of the nitrous acid on the amide, and that ammonia and the organic acid are formed, the ammonia, however, *in statu nascenti*, becoming decomposed with the nitrous acid into water, and nitrogen; thus, for instance, acetamide and nitrous acid yield water, acetic acid, and nitrogen, for  $C_4H_5NO_2 + NO_3 = C_4H_3O_3 + 2HO + 2N$ . In this way we may hope that several nitrogenous animal matters may be discovered to be amides, as in the case of asparagin, which has been shown to be the amide of malic acid.

If the amides of these acids be treated with anhydrous phosphoric acid, they lose 2 atoms of water, and nitrogenous bodies rich in oxygen remain, which contain the radical of the acid and have 1 equiv. of nitrogen in place of the 3 atoms of oxygen. These bodies have been named *nitriles*. Notwithstanding the similarity of their composition with that of the volatile oxygenous alkaloids, they possess no basic properties.

Valeramide and phosphoric acid form hydrated phosphoric acid and valeronitrile:  $C_{10}H_{11}NO_2 + PO_5 = PO_5.2HO + C_{10}H_9N$ .

The amides of this group are finally distinguished by a property which is not common to the amides of most other acids; when treated with potassium they yield cyanide of potassium and a carbohydrogen. Hence it seems probable that cyanogen exists pre-formed in these amides, since, from their total want of basic properties, it cannot be supposed that they contain a conjugated ammonia and that 1 atom of oxygen can be replaced by amide.

Taking this view, acetamide must be regarded as hydrocyanate of wood-spirit, and metacetamide as hydrocyanate of alcohol, for  $C_4H_5NO_2 = C_2H_4O_2.HC_2N$ , and  $C_6H_7NO_2 = C_4H_6O_2.HC_2N$ .

The amides lead us at once to a further consideration of the *nitriles*, which are equally important in reference to our knowledge of the arrangement of atoms and the metamorphosis of matter.

These bodies are, in part, formed during the decomposition of animal substances by oxidizing agents; they may, however, be obtained by treating the corresponding ammonia-salt or the amide with anhydrous phosphoric acid. This mode of preparation is especially applicable for the nitriles of this group of acids; others are prepared either by the mere exposure of the ammonia-salt to heat, or by passing the vapor over heated caustic lime.

<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. t. 22, pp. 170-179.

The nitriles are oily, very volatile fluids, less soluble in water than in alcohol and ether, and having a peculiar odor; they can be distilled without undergoing decomposition, have no action on vegetable colors, and do not unite with acids to form salts. They unite, however, directly with sulphuretted hydrogen, assimilating 2 equivalents of it, so that sulphurous substances analogous to the amides are produced; thus, for instance, benzonitrile, with sulphuretted hydrogen, forms sulphobenzamide, which is analogous to benzamide:  $C_{14}H_5N + 2HS = C_{14}H_7NS_2 \sim C_{14}H_7NO_2$ .

Alkalies and strong acids reduce most of the nitriles to their original component parts, that is to say, to ammonia and the corresponding acid, by assimilating 3 atoms of water; thus, for instance, in the case of valeritrile:  $C_{10}H_9N + 3HO = H_3N + C_{10}H_9O_3$ .

Several of the properties of the nitriles, and especially the modes in which they are decomposed, indicate that in their chemical constitution they are not to be regarded as compounds of the radical of the corresponding acid with nitrogen, but rather as combinations of cyanogen and certain carbo-hydrogens;—a view which throws a perfectly new light on the theoretical composition of the acids of this group.

If we first glance at the nitriles of the simplest acids of this group,—those of formic acid, acetic acid, and metacetic acid,—it becomes manifest that these are bodies which have been long known, but never have been, nor can be, regarded as nitriles. The nitrile of formic acid must be  $C_2HN$ ; this, however, is the composition of hydrocyanic acid, which, as is well known, is also obtained by heating formate of ammonia, three atoms of water being separated. Hydrocyanic acid can, however, as we know, be readily converted, like the nitriles, into ammonia and the corresponding (formic) acid.

If, farther, with the view of preparing the nitrile of acetic acid, acetamide be mixed with anhydrous phosphoric acid, another long-known body, supposed to be otherwise constituted, is formed, namely, cyanide of methyl, for  $C_4H_3N = C_2H_3.C_2N$ . The nitrile of metacetic acid which corresponds to cyanide of ethyl, behaves in a perfectly similar manner, for  $C_6H_5N = C_4H_5.C_2N$ . An intelligent observer, Kolbe,<sup>1</sup> who has instituted very excellent observations on the subject, struck upon the idea of preparing metacetic acid from the cyanide of ethyl (obtained by the distillation of sulphate of oxide of ethyl and potash, and cyanide of potassium), by treating it with solution of potash; and the attempt completely succeeded, for the cyanide of ethyl (perfectly corresponding in its nature to the aforesaid nitrile), took up 3 atoms of water, and became decomposed into ammonia and metacetic acid, according to the formula,  $C_4H_5.C_2N + 3HO = H_3N + C_6H_5O_3$ .

From these facts he was led to regard the nitriles (as far as they are yet known) of the acids of this group as combinations of cyanogen with a radical of the haloid bases pertaining to the ether group, that is to say, with a carbo-hydrogen in which there are contained a large number of atoms of carbon, and the next higher odd number of atoms of hydrogen.

<sup>1</sup> Phil. Mag. Vol. 31, pp. 266-271.

Thus, these substances arrange themselves in the following arithmetical proportion :—

Nitrile of formic acid, . . . . .	= hydrocyanic acid, . . . . .	= $\text{H.C}_2\text{N}$ .
— — — acetic acid, . . . . .	= cyanide of methyl, . . . . .	= $\text{C}_2\text{H}_3.\text{C}_2\text{N}$ .
— — — metacetic acid, . . . . .	= cyanide of ethyl, . . . . .	= $\text{C}_4\text{H}_5.\text{C}_2\text{N}$ .
Butyronitrile, . . . . .		= $\text{C}_6\text{H}_7.\text{C}_2\text{N}$ .
Valeronitrile, . . . . .		= $\text{C}_8\text{H}_9.\text{C}_2\text{N}$ .

While in the first three of these combinations the existence of cyanogen may be regarded as established, Kolbe<sup>1</sup> believed that he could recognize the existence of such carbo-hydrogens as  $\text{C}_6\text{H}_7$  and  $\text{C}_8\text{H}_9$ ; and, indeed, he fully proved their presence, by exposing to an electric current the potash-salts of the acids corresponding to the two last-named nitriles, namely, butyric acid and valerianic acid; besides other products, he then obtained the carbo-hydrogens  $\text{C}_6\text{H}_7$  and  $\text{C}_8\text{H}_9$ . In further investigations,<sup>2</sup> by decomposing cyanide of ethyl by potassium, he established the existence of the radicals, methyl and ethyl,  $\text{C}_2\text{H}_3$  and  $\text{C}_4\text{H}_5$ .

From these facts relating to the nitriles of these acids, we are almost involuntarily led to Kolbe's original view, and to regard the acids of this group as conjugated oxalic acids, that is to say, as acids in which oxalic acid is so combined with one of the above-named carbo-hydrogens =  $\text{C}_n\text{H}_{n+1}$ , as not to affect the saturating capacity of the acid.

This view is supported by the following experimental evidence.

Butyric and valerianic acids are decomposed under the influence of the galvanic current; assimilating an atom of oxygen, they yield 2 equivs. of carbonic acid and the corresponding carbo-hydrogen.

Cyanogen with water becomes decomposed, as is well known, into oxalic acid and ammonia ( $\text{C}_2\text{N} + 3\text{H}_2\text{O} = \text{H}_2\text{N} + \text{C}_2\text{O}_3$ ); conversely, on heating oxalate of ammonia, cyanogen, together with oxamide, is formed. The production and decomposition of valeronitrile may hence be explained in the following manner: if valerianic acid be an oxalic acid conjugated with the carbo-hydrogen; *valyl* =  $\text{C}_8\text{H}_9$ , the latter is converted into cyanogen by the metamorphosis of the ammonia-salt into nitrile; and the cyanogen combining with the adjunct  $\text{C}_8\text{H}_9$ , yields the empirical formula for valeronitrile. If, however, the latter be regarded as cyanide of valyl, and be decomposed by alkalies, the conjugated cyanogen, just as if it were isolated, becomes converted into ammonia and oxalic acid, which then remains in combination with the adjunct  $\text{C}_8\text{H}_9$ .

Considering the subject in this point of view, we must regard the acids of this group as constituted in the following manner :—

Formic acid, . . . . .	= hydrogen-oxalic acid, . . . . .	= $\text{H.C}_2\text{O}_3$ .
Acetic acid, . . . . .	= methyloxalic acid, . . . . .	= $\text{C}_2\text{H}_3.\text{C}_2\text{O}_3$ .
Metacetic acid, . . . . .	= ethyloxalic acid, . . . . .	= $\text{C}_4\text{H}_5.\text{C}_2\text{O}_3$ .
Butyric acid, . . . . .	= metethyloxalic acid, . . . . .	= $\text{C}_6\text{H}_7.\text{C}_2\text{O}_3$ .
Valerianic acid, . . . . .	= valyloxalic acid, . . . . .	= $\text{C}_8\text{H}_9.\text{C}_2\text{O}_3$ .
Caproic acid, . . . . .	= amyloxalic acid, . . . . .	= $\text{C}_{10}\text{H}_{11}.\text{C}_2\text{O}_3$ .

Closely allied to this view of the constitution of these acids is another consideration, which has reference to the production of these homologous

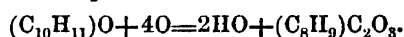
<sup>1</sup> Chem. Gaz. Vol. 5, p. 228.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 271-288.

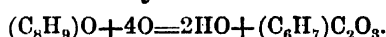


acids from the series of the ether-like, homologous haloid bases. The general formula of the haloid bases,—oxide of methyl, oxide of ethyl, and oxide of amyl, is  $=C_nH_{n+1}O$ , while the formula of the acids is  $C_nH_{n-1}O_3$ ; we have explained the production of the acids from the corresponding haloid bases by the simple assimilation of 4 atoms of oxygen, and loss of 2 atoms of water; as, for instance, in the conversion of oxide of ethyl into acetic acid: if, however, the above conclusions, which have been derived from simple inductions, be correct, it must be assumed that (to take a definite case) in the conversion of oxide of ethyl into acetic acid, the complex atom,  $C_2H_5$ , leaves the radical of the oxide of ethyl,  $C_4H_5O$ , and unites with 4 extraneous atoms of oxygen, and with the 1 atom which is presented in oxide of ethyl, to form water and oxalic acid, which combines with the radical of the next lower haloid base, methyl, and represents acetic acid.

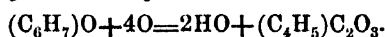
Oxide of amyl yields valyloxalic acid:



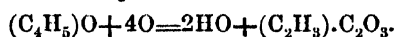
Oxide of valyl yields metethyloxalic acid:



Oxide of metethyl yields ethyloxalic acid:



Oxide of ethyl yields methyloxalic acid:



As, according to this view, oxalic acid constitutes the acidifying principle of the bodies of this group, we shall consider it the first in the series of acids.

### OXALIC ACID.— $C_2O_3.HO$ .

#### *Chemical Relations.*

*Properties.*—This acid crystallizes with 3 atoms of water in oblique rhombic prisms, is devoid of smell, has a sharp acid taste, and effloresces on exposure to the air, losing 2 atoms of water and becoming disintegrated into a white powder; on heating it carefully to  $150^\circ$  or  $160^\circ$ , it sublimes undecomposed in acicular crystals; but at  $170^\circ$  (or if the crystallized acid be rapidly heated to  $155^\circ$ ) it becomes decomposed into carbonic oxide and carbonic acid, a little formic acid and water; it dissolves in 8 parts of cold and 1 part of boiling water, and in 4 parts of spirit of wine; its solutions redden litmus strongly. On boiling oxalic acid with solution of oxide or chloride of gold, carbonic acid is evolved, and the gold is precipitated in the form of extremely fine black powder. Treated with concentrated sulphuric acid, it becomes decomposed into carbonic oxide and carbonic acid, and effects no change in the color of the sulphuric acid.

*Composition.*—In accordance with the above formula, this acid, which cannot exist in the free state without water, contains in 100 parts :

Carbon,	.	.	.	.	.	2 atoms	=	26.667
Hydrogen,	.	.	.	.	.	3 "	=	58.833
Water,	.	.	.	.	.	1 "	=	20.000
								<hr/>
								100.000

The atomic weight of the hypothetical anhydrous acid=450.0; its saturating capacity=22.222.

In reference to the history of this acid, we may observe that while some chemists regard it as the oxide of an oxygenous radical, oxalyl= $C_2O_2$ , in consequence of the preponderance of its acidity over that of carbonic acid, others regard it as a hydrogen acid= $C_2O_3.H$ .

*Combinations.*—Oxalic acid combines with alkalies in three proportions, in which the oxygen of the base is to that of the acid as 1 : 3, 1 : 6, and 1 : 12 respectively. These salts are soluble in water, but all other oxalates are insoluble, or only very slightly soluble, in that fluid: none of the oxalates are soluble in alcohol. These salts do not char when heated. The combinations of oxalic acid with the more easily reducible oxides, yield carbonic acid and the reduced metal (thus, for instance,  $CoO.C_2O_3=2CO_2+Co$ ); while those with less easily reducible bases evolve carbonic oxide gas, and are converted into carbonates.

*Oxalate of Ammonia*, neutral oxalate of oxide of ammonium,  $H_4NO.C_2O_3+2H_2O$ , is obtained by neutralizing oxalic acid with carbonate of ammonia, and evaporating the solution; it crystallizes in needles, has a saline taste, effloresces on exposure to the atmosphere, and its solubility in water is less than that of oxalic acid.

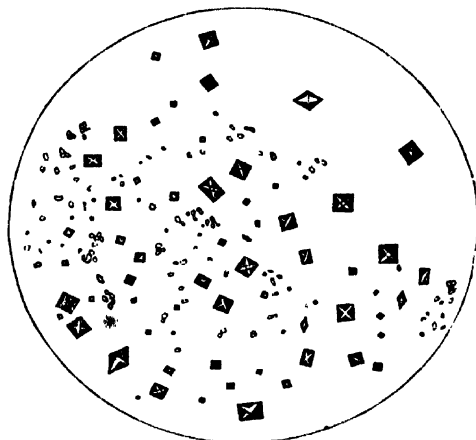
*Oxamide*,  $C_2H_2NO_2 (=H_2N.C_2O_2)$  is obtained either by the dry distillation of oxalate of ammonia, or by the treatment of neutral oxalate of oxide of ethyl with ammonia; it has a crystalline powdery appearance, is of a glistening white color, has no smell or taste, and dissolves very slightly in cold, but rather more freely in hot water; when strongly heated it becomes decomposed into water, carbonic oxide, hydrocyanic acid, and a little urea. If a sufficient quantity of water be present, a very small quantity of oxalic acid can convert an infinite quantity of oxamide into oxalate of ammonia.

*Oxamic Acid*,  $C_4H_2NO_5.HO$ , is an acid in which we assume that oxalic acid is conjugated with oxamide ( $C_2H_2NO_2.C_2O_3.HO$ ); it is produced by the dry distillation of binoxalate of ammonia; it occurs as a colorless, granular, inodorous powder, which is not readily soluble in water, and reddens litmus. When heated with sulphuric acid it becomes decomposed into ammonia and oxalic acid; its salts are for the most part soluble; at least its baryta, lime, and silver salts dissolve in boiling water.

*Oxalate of Lime*,  $CaO.C_2O_3$ , is a very important substance in pathological chemistry; it occurs as a white, tasteless, and inodorous powder, which, however, under the microscope, is found to exhibit a distinct crystalline form. These crystals, whose crystallographic relations have

been carefully studied by C. Schmidt,<sup>1</sup> appear, when seen with a low power, as envelope-formed, sharply defined bodies; but when more highly magnified, they may easily be recognized as obtuse square octohedra (Fig. 1;) some, however, among them, are very acute. These crystals con-

Fig. 1.



Crystals of Oxalate of Lime.

tain 1 atom of water, which they lose at  $180^{\circ}$ . Oxalate of lime is all but insoluble in water, and it is almost proof against the action of acetic and oxalic acids; it readily dissolves, however, in the stronger mineral acids.

Artificially prepared oxalate of lime only shows these crystals, when very dilute solutions of salts of lime have been mixed with diluted boiling solutions of alkaline oxalates; under other circumstances it appears under the microscope merely in spherical or nodular masses. Crystals of oxalate of lime may be distinguished from those of chloride of sodium, which they much resemble in form, by the easy solubility of the latter in water, and by their transparency. Larger crystals of oxalate of lime sometimes occur, having some resemblance to crystals of phosphate of ammonia-magnesia, which in the projection resemble a square octahedron; but a more accurate microscopic examination and the solubility of the triple phosphate in acetic acid enable us to discriminate between these crystals and those of oxalate of lime. Golding Bird<sup>2</sup> also describes crystals of oxalate of lime shaped like dumb-bells or rather like two kidneys with their concavities opposed, and sometimes so closely approximating as to appear circular, the surface being finely striated. These crystals are produced, in all probability, by a zeolitic arrangement of minute acicular crystals presenting a physical structure resembling that of spherical crystals of carbonate of lime. [Dr. Golding Bird<sup>3</sup> has recently shown that in all probability these dumb-bell crystals consist of *oxalurate of lime*.—G. E. D.]

<sup>1</sup> Entwurf einer allg. Untersuchungsmethode der Säfte und Excrete des thierischen Organismus. Mitau u. Leipz. 1846, S. 63-65.

<sup>2</sup> Urinary Deposits; their diagnosis, pathology, and therapeutical indications. Am. edition, p. 184.

<sup>3</sup> Op. cit. p. 187.

Other oxalates have at present excited no physiological interest.

*Preparation.*—Oxalic acid is a final product of the oxidation of most animal and vegetable bodies; hence it may be prepared from very different substances by strong oxidizing agents: it is most commonly obtained by the decomposition of sugar by not too concentrated nitric acid, by evaporation to crystallization, and finally by recrystallization in water.

*Tests.*—Oxalic acid and its salts are so well characterized that it is hardly possible to mistake them for any other bodies. In the animal organism oxalic acid is almost always combined with lime, and with a little practice this salt may be readily discovered by the microscope, and by the insolubility of its crystals in acetic acid. Should a further investigation appear necessary, the presence of oxalic acid might be determined by its property of reducing gold from its solutions, and by its not charring either in the free or in the combined state when heated, or on the application of sulphuric acid. Oxalate of lime can be separated from most of the substances with which it is likely to be mixed either by acetic acid or by dilute solution of potash.

### *Physiological Relations.*

*Occurrence.*—Frequently as oxalic acid, combined either with the alkalies or with lime, occurs in the vegetable kingdom (Schleiden,<sup>1</sup> Carl Schmidt,<sup>2</sup> and others), it is very seldom found in the animal organism, at least in large quantities. It only occurs in the latter in combination with lime, never being present in sufficient quantity to combine with the alkalies as well as with lime. Moreover, it is much more frequently met with in pathological than in physiological conditions.

It is in the urine that the presence of oxalate of lime has been most frequently observed; it was for a long time regarded as a morbid product in this fluid, but independently of the circumstance that this body is constantly present, together with carbonate of lime, in the urine of herbivorous animals, it has frequently been found in normal human urine by myself,<sup>3</sup> Höfle,<sup>4</sup> and others.

In examining microscopically the morning urine of healthy men I have frequently discovered isolated crystals of oxalate of lime; this is not, however, always the case; and further, the oxalate of lime recognizable in such cases by the microscope is not all that is contained in the urine, for it forms in larger quantities after some time, and during the acid urinary fermentation so admirably described by Scherer. We must not forget that oxalate of lime may possibly be formed during this process. We know that there is a close connection between the excretion of uric acid and the formation of this salt, from the circumstance that in most specimens of urine, both sedimentary and non-sedimentary, oxalate of lime cannot be recognized by the microscope so long as the fluid is fresh, but as soon as crystals of uric acid present themselves, crystals of oxalate of lime (at all events in small numbers) may also be discovered; indeed, we generally find that in morbid urine the abundance

<sup>1</sup> Grundzüge der Botanik. 2 Aufl. 1846.

<sup>2</sup> Entwurf u. s. w.

<sup>3</sup> Wagner's Handwörterbuch der Physiologie. Bd. 2, S. 6.

<sup>4</sup> Chemie und Mikroskop am Krankenbette. Erlangen, 1848. S. 385.

of these crystals is proportional to the rapidity with which the free uric acid separates. Since uric acid, when acted upon by certain oxidizing agents, may be decomposed into urea, allantoin, and oxalic acid, we may assume that a portion of the uric acid may be decomposed during this acid urinary fermentation, and that oxalic acid is formed from it—a possibility which is converted into a probability by the recent observation of Ranke,<sup>1</sup> that uric acid, on the addition of yeast and of an alkali, becomes decomposed at a high temperature into urea and oxalic acid. After allowing morning urine to stand for a considerable time, we often find a great many of these crystals, when the perfectly fresh urine presented no trace of them. The following is an excellent mode of demonstrating the existence of oxalate of lime in normal urine. If it be winter we must expose fresh urine out of doors till it freezes; in this process, as in the freezing of wine and vinegar, a great part of the water crystallizes in a comparatively pure state, and after its removal we obtain a concentrated saline solution in which microscopic crystals of oxalate of lime may be discovered. That oxalate of lime is at first actually held in solution in filtered urine, and that it does not, as C. Schmidt supposes, proceed from the mucus of the bladder, is a view which is supported by the experiment which I have often repeated, that in urine, which after thoroughly cooling was freed from its mucus and urate of soda by filtration, the most distinct crystals of oxalate of lime might after a time be recognized, while no traces of them could either previously be detected in the mucus of the fresh urine, or found after the residue on the filter had been for some time in contact with water. The oxalate of lime, with a few crystals of uric acid, does not separate from filtered urine until after it has stood for some time. We may very easily convince ourselves that oxalate of lime is present in a state of solution, by extracting the solid residue of filtered urine with not too concentrated spirit, and agitating the spirituous extract with ether; after the extraction with ether, there may be observed, in the alcoholic extract, a sediment insoluble in water, which consists of the most beautiful crystals of this salt. While in the acid urinary fermentation the separation of the oxalate of lime increases with the augmentation of the free acid of the urine, in the latter case the salt is separated by the removal of the free acid.

The quantity of oxalate of lime in ordinary urine is so minute, that, till recently, chemists, from the want of sufficiently accurate means of analysis, were unable to recognize it; good analysts have, however, always found, in the insoluble part of the ash of the extract of urine, a little carbonate of lime, which, at all events, owes part of its origin to the oxalate of lime.

Crystals of oxalate of lime are most frequently found in the urine after the use of vegetable food, especially of such kinds as contain ready formed oxalates (Wilson).<sup>2</sup> Donné found that after the use of sparkling wines, the quantity of the salt is increased in the urine; and my own experiments show that there is an increased secretion of oxalate of lime

<sup>1</sup> Journ. f. pr. Ch. Bd. 56, S. 16.

<sup>2</sup> Provincial Medical and Surgical Journal, 1846, p. 413.

after the use of beer containing much carbonic acid and of the alkaline bicarbonates and vegetable salts. I cannot confirm Bird's view that highly nitrogenous food causes a precipitate or even an augmentation of the oxalate of lime. It is often found in the urine of pregnant women (Höfle).<sup>1</sup>

From a series of direct experiments on the subject, C. Schmidt<sup>2</sup> is led to deny that oxalate of lime introduced into the stomach, passes into the urine; and in this point I can perfectly confirm him, without, however, going so far as to assert that the food exerts no influence on the formation of this body. In the excrements of caterpillars we often find much oxalate of lime which is not formed directly from the ingesta, since I<sup>3</sup> have very often found the crystals in the biliary ducts of these animals. Preparations can be easily made of these organs, and in consequence of their contractility a large quantity of their contents may be expressed from the cut tubes, and submitted to microscopic examination.

With reference to the occurrence of oxalate of lime in certain morbid conditions, Prout, Bird, and others, make very different statements, none of which are yet fully established. Numerous examinations of morbid urine have convinced me, that in this country, at least, the sediments of oxalate of lime are much rarer than they are represented to be by English writers. These investigations have led me to the following results; when the respiratory process is in any way disturbed, we most frequently observe a copious excretion of oxalate of lime; it is most common either in fully developed pulmonary emphysema, or when the pulmonary tissue has lost much of its elasticity after repeated catarrhs; on the other hand, it is not present nearly so often in inflammatory or tuberculous affections of the lungs (Höfle);<sup>4</sup> moreover, it is common in convalescence from severe diseases, as for instance, typhus, mucus-corpuscles being then often associated with a trifling sediment of oxalate of lime. [The frequent occurrence of oxalate of lime in the urine during convalescence has been independently observed by Professor Walsh. See his paper on the oxalates in the *Monthly Journal of Medical Science*, Jan. 1849. G. E. D.] I have only met with actually pure sediments of this salt in three persons, who, sometimes (at somewhat considerable intervals), suffered from epileptic attacks. It is by no means constant, according to my experience, in the urine of rachitic children (Simon),<sup>5</sup> of gouty adults with osteoporosis, of women with leucorrhœa, of patients with heart-disease, or in urine containing semen (Donné).<sup>6</sup>

In the dyspeptic conditions in which Prout and Bird have found sediments of oxalate of lime, I have failed in discovering anything of the sort; on the contrary, I have generally found the sediments in the urine of such patients to be free from these crystals. The reason why the English have so often found this salt in the urine, may be, that in England (as we shall further notice at a future page), the urine is generally

<sup>1</sup> Chemie u. Mikroskop u. s. w. S. 385.

<sup>2</sup> Entwurf u. s. w. S. 70.

<sup>3</sup> Jahresbericht d. ges. Med. 1844. S. 25.

<sup>4</sup> Chemie u. Mikroskop u. s. w. Nachtrag, S. 176.

<sup>5</sup> Hufeland's Journal, 1841. Dec S. 73-88.

<sup>6</sup> Cours de microscopie. pp. 249, 322.

in a more concentrated state than in Germany, and as Bird very correctly remarks, oxalate of lime is more rapidly separated from a concentrated than an aqueous urine. Moreover, experience at the bedside teaches every unprejudiced observer that the appearance of oxalate of lime in the urine is by no means accompanied by the group of symptoms which certain English physicians describe as pertaining to what they call the oxalic diathesis. [For the arguments in opposition to this opinion the reader is referred to Dr. Golding Bird's *Urinary Deposits*, 3d ed. p. 230.—G. E. D.]

That the mulberry calculus consists for the most part of oxalate of lime, has been long known; but most other urinary calculi, whether they consist principally of earths or urates, almost always contain a little oxalate of lime.

This salt has only rarely been found in other places besides the urine. C. Schmidt has remarked that it is often present in the mucus of the gall-bladder, and that it is scarcely ever absent from the mucous membrane of the impregnated uterus. I once discovered oxalate of lime in expectorated matter, but whether it was produced from the pulmonary mucus, or from fragments of food in the mouth, I could not decide. [Dr. Garrod<sup>1</sup> has recently detected oxalic acid in the blood in a case of chronic hiccup and vomiting, and in several cases of gout.—G. E. D.]

*Origin.*—As the use of vegetable food, of which many varieties contain oxalates, increases the quantity of oxalate of lime in the urine, the inference would seem a legitimate one, that the oxalates are transmitted from the food to the urine. The source of this salt must, however, not be sought for only in the pre-formed oxalates, but in the amount of alkalies in combination with vegetable acids present in the food; for, as we have already mentioned, they induce an augmentation of the oxalate of lime. In all the well-marked cases to which I have alluded, the increase of the oxalate of lime seemed to be combined with disturbance of the respiratory process. Thus it may easily be understood why, after the use of drinks rich in carbonic acid, of alkaline bicarbonates, or vegetable salts, oxalic acid is increased in the urine; the superfluous carbonic acid which has entered the blood, or been generated there from the salts of organic acids, must obstruct the absorption of oxygen and the perfect oxidation of certain substances in the blood; hence also the quantity of oxalate of lime has been found to be increased by the partially impeded exchange of oxygen and carbonic acid in the lungs, consequent on emphysema, pulmonary compression during pregnancy, &c. We might, in such cases, assume, according to a formerly prevalent belief, that the kidneys in some degree acted vicariously for the lungs, since under the form of oxalic acid they remove from the organism the carbon which the latter organs would have excreted as carbonic acid.

Although certain chemists hold a contrary opinion, it is an undoubted fact that the nervous system has an influence on the oxidation of the blood. The occurrence of oxalate of lime in cases of epileptic convulsions, in convalescent persons, &c., might be referred to the disturbance

<sup>1</sup> Medico-chirurgical Transactions. Vol. 82, p. 171.

induced in such cases in the nutrition or in the function of the nervous system, and to its diminished influence on the process of respiration, without there being any necessity for the assumption of a special diathesis.

It seems, moreover, unreasonable to set up such a diathesis, since the establishment of a special disease from a single symptom—that symptom being only the occurrence of oxalate of lime—is entirely opposed to the spirit of rational medicine.

From Wöhler and Liebig's discovery that uric acid is decomposed by peroxide of lead into urea, allantoin, and oxalic acid, it has been pretty generally assumed that the oxalic acid of the urine is due to an oxidation of the uric acid; the oxalic acid, in this case, not being converted into carbonic acid, as usually occurs in the healthy organism. Wöhler and Frerichs<sup>1</sup> have since shown by direct experiments that uric acid is decomposed in the animal organism in precisely the same way as by peroxide of lead, since they found that after the injection of urates there was not merely an augmentation of the urea in the urine, but also that oxalic acid was present in it in larger quantity. That the formation of oxalic acid may be in part thus explained, is unquestionable, but there are many other substances in the animal organism besides uric acid, which by oxidation yield oxalic acid. No definite numerical ratio between the uric acid, urea, and oxalate of lime in the urine, has been yet established.

C. Schmidt<sup>2</sup> has propounded a very ingenious view regarding the origin of oxalate of lime in the urine. He believes that we must seek for the source of its secretion in the mucous membrane of the urinary passages, and that the oxalate of lime is first produced by the decomposing action of the acid urine on a soluble compound, oxalate of albumen-lime, secreted by the mucous membranes; for oxalate of lime as an insoluble body could not penetrate with the urine through a series of renal cells: oxalate of lime is also formed from the mucus of the gall-bladder by this mode of decomposition. When oxalate of lime occurs in the urine, we always find an augmentation of the mucus. These reasons do not, however, appear to be so decisive as to induce us to exchange the view we have already given for that of Schmidt; and indeed in another place we find Schmidt<sup>3</sup> himself maintaining that the urea is in part combined with oxalic acid.

## FORMIC ACID.— $C_2HO_3.HO$ .

### *Chemical Relations.*

*Properties.*—This acid possesses the general characters of the acids of this group; with water it forms two distinct hydrates, one of which becomes solid at  $-1^\circ$ , boils at  $+99^\circ$ , and has a specific gravity of 1.2353, while the other, which contains 48.35% or 2 atoms of water, does

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 340.

<sup>2</sup> Ib. Bd. 60, S. 55, ff.

<sup>3</sup> Entwurf u. s. w. S. 47.



not solidify at a temperature of  $-15^{\circ}$ , boils at  $+106^{\circ}$ , and has a specific gravity of 1.1104. By concentrated sulphuric acid it is decomposed into water and carbonic oxide ( $C_2H_2O_3 = HO + 2CO$ ); the salts of oxide of silver and of oxide of mercury are reduced when warmed in it.

*Composition.*—In correspondence with the above formula, 100 parts of this acid must contain :

Carbon,	.	.	.	.	2 atoms,	.	.	.	.	26.087
Hydrogen,	.	.	.	.	1 "	.	.	.	.	2.174
Oxygen,	.	.	.	.	3 "	.	.	.	.	52.174
Water,	.	.	.	.	1 "	.	.	.	.	19.565
										<hr/> 100.000

The atomic weight of the hypothetical anhydrous acid = 462.5; its saturating capacity = 21.62. According to the theory which we have laid down, formic acid should be regarded as an oxalic acid conjugated with hydrogen =  $H.C_2O_3 + HIO$ ; but according to ordinary views it is assumed to contain a radical *formyl* =  $C_2H$ , which is believed to occur in several other combinations, as for instance in chloroform.

*Combinations.*—The salts of formic acid are soluble; with alkalis, it also forms acid salts.

*Formate of ammonia* is known by its property of becoming converted on heating into hydrocyanic acid ( $H_4NO.C_2H_2O_3 = H.C_2N + 4HIO$ ), and hence the hydrocyanic acid which often appears during the decomposition of animal substances may be dependent on the previous formation of formate of ammonia.

There are certain combinations, which in reference to their empirical composition, may be regarded as formic acid, but in which the whole of the oxygen is replaced by chlorine, bromine, iodine, or sulphur; the best known of these is *chloroform* or *perchloride of formyl*,  $C_2HCl_3$ , which is employed in place of ether to induce anæsthesia.

*Preparation.*—This acid was most commonly obtained in former times by distilling a large quantity of ants with water or spirit: from the distillate, which naturally only contained the acid in a very dilute state, the concentrated acid was obtained according to the ordinary methods by saturation with a base, and by the decomposition of the crystallized salt with sulphuric acid. As, however, we have since ascertained that formic acid is a product of the oxidation of many animal and vegetable substances, we are now in the habit of obtaining it from various sources by the action of oxidizing agents, as peroxide of manganese and sulphuric acid, chromic acid, or hypermanganic acid. It is best obtained by adding a little water and sulphuric acid to a mixture of three parts of sugar and one part of bichromate of potash (2 atoms of  $SO_3$  to 1 atom of  $KO.2CrO_3$ ) and by distilling.

*Tests.*—This acid may be readily distinguished from most other acids by its volatility, and from other acids of this group by its power of reducing the oxides of mercury and of silver; but it must be recollected that if we obtain formic acid by the distillation of a mixture with sulphuric acid, this formic acid may have been produced by the action of the sulphuric acid on organic matter, or on already formed hydrocyanic acid. We may separate it from the other acids of this group by

fractional distillation, since the boiling-point of this acid is lower than that of all other homologous acids.

### *Physiological Relations.*

*Occurrence.*—Formic acid has hitherto been much more frequently found as a product of the decomposition of many organic substances, as for instance in the gradual decay (Eremacausis) of coal, than as an educt of the animal body. It has only as yet been positively proved to exist pre-formed in ants (especially *Formica rufa*); Bouchardat and Sandras<sup>1</sup> believe, however, that they have found it in the blood of dogs which for a long time had been fed with sugar. According to Scherer,<sup>2</sup> there are contained in the juice of flesh not only lactic, inosic, and phosphoric acids, but also formic, acetic, and several other acids of this group.

Scherer has likewise found formic acid, in association with other acids of this group, in the acid fluid of the spleen<sup>3</sup> and in leucæmic blood.<sup>4</sup> Further, very large quantities of this acid have been obtained, under my own inspection, from normal human sweat, the exact nature of the acid being not only determined by the reactions described, but also by the determination of its saturating capacity and by elementary analysis.

[Will of Erlangen has recently shown that the active poisonous principle in certain caterpillars is formic acid. It exists in a free, concentrated state in all parts of the animal, particularly in the fæces, in the greenish-yellow matter that exudes when the animal is cut, and in the hollow bristles.—G. E. D.]

*Origin.*—Notwithstanding that the principal processes in the animal organism are based on an oxidation, and that, on the other hand, in the artificial oxidations of animal substances, formic acid is produced, we do but rarely meet with this acid in the animal kingdom: indeed, even with reference to the ants, it is by no means certain that they actually produce formic acid, for we know that juniper berries and the cones of several kinds of pine contain formic acid, and that these substances are much sought after by ants. We must leave this question unanswered, since it is only by direct experiments that we can determine whether ants take up exactly the same amount of acid as they yield.

Bouchardat and Sandras are of opinion that the lactic acid formed from starch and sugar in the blood is first decomposed into formic acid before its elements are finally reduced to water and carbonic acid.

### ACETIC ACID.— $C_4H_3O_3.HO$ .

#### *Chemical Relations.*

*Properties.*—Acetic acid has the general characters of the acids of this group. In its most concentrated state, as first hydrate, it forms a

<sup>1</sup> Compt. rend. T. 20, pp. 1026 et 1085.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 69, S. 196–201.

<sup>3</sup> Verhandl. d. phys.-med. in Ges. Würzb. Bd. 2, S. 298.

<sup>4</sup> Ibid. p. 321.

crystalline mass below  $+16^{\circ}$ ; above this temperature it is fluid, has a specific gravity of 1.080, and boils at  $117.3^{\circ}$ ; its second hydrate, containing 2 atoms of water, has a specific gravity of 1.078 and boils at  $140^{\circ}$ .

We shall notice only the most important points regarding acetic acid and its compounds, and those having an especial bearing on animal chemistry; the other compounds of acetic acid pertaining to pure rather than to physiological chemistry.

*Composition.*—According to the above formula, acetic acid consists of:

Carbon,	.	.	.	.	4 atoms,	.	.	.	.	40.000
Hydrogen,	.	.	.	.	3 "	.	.	.	.	5.000
Oxygen,	.	.	.	.	3 "	.	.	.	.	40.000
Water,	.	.	.	.	1 "	.	.	.	.	15.000
										<hr/>
										100.000

The atomic weight of the hypothetical anhydrous acid = 637.5; its saturating capacity = 15.686. Kolbe's hypothesis that acetic acid is oxalic acid conjugated with methyl =  $C_2H_3.C_2O_3.HO$ , was anticipated by Berzelius. Till then it was assumed that the radical  $C_4H_3$  existed in acetic acid, and aldehyde and aldehydic acid were regarded as lower stages of oxidation of the same radical.

*Combinations.*—The only acid acetate with which we are acquainted is a potash-salt; with the oxides of the heavy metals it has a strong tendency to form basic salts.

*Acetamide*,  $H_2N.C_4H_3O_2=C_4H_5NO_2$ , is prepared from acetic ether and ammonia; it forms a white, crystalline, diffuent mass, which fuses at  $78^{\circ}$  and boils at  $228^{\circ}$ ; it has a sweetish, cooling taste; by anhydrous phosphoric acid it is converted into cyanide of methyl; hence it has been considered as hydrocyanate of wood-spirit ( $C_4H_5NO_2=C_2H_3O+HC_2N+HO$ ).

By dry distillation of the acetates with strong bases, we obtain *acetone* or *hydrated oxide of anyl*,  $C_6H_5O.HO$ , which presents much similarity with the alcohols of the haloid bases.

On heating equal parts of acetate of potash and arsenious acid in a retort, we obtain alkarsin or oxide of kakodyl,  $C_4H_5As_2O$ , which is distinguished by its very specific odor.

*Preparation.*—The methods of producing and obtaining acetic acid are so well known that we need not here advert to them.

*Tests.*—Some light will be thrown on the importance of the modes of testing for acetic acid when we have to treat of the assumed or actual occurrence of acetic acid in the animal fluids.

As in the case of most organic substances, we must first separate it from most of the substances with which it is mixed, before we can apply the appropriate tests. This separation is comparatively easy, because the acid admits of being distilled; hence it can only be confounded with volatile acids exhibiting reactions homologous or similar to it. It may be readily distinguished from formic acid, in consequence of the property which this latter acid possesses of being decomposed by oxide of mercury;

hence these two acids can hardly be mistaken for one another. How it is to be separated and distinguished from the homologous acids, as, for instance, metacetic acid, &c., will be explained when we treat of these acids.

If we have isolated acetic acid as completely as possible by distillation, and then by crystallization of one of its salts, the following reactions may be established, independently of the examination of the form of the crystals; nitrate of suboxide of mercury added to a not too dilute solution of an acetate at first yields no precipitate, but, after a short time, minute crystalline specks are formed, which slowly gravitate in the fluid like fatty glistening scales. Since the acetates, in common with the meconates and sulphocyanides, yield a somewhat intense red color on the addition of a solution of a persalt of iron, acetic acid, in a mixed fluid, might be mistaken for one of these acids; but acetic acid may be readily distinguished from meconic acid by the solubility of the acetate of lime (the meconate of lime being insoluble in water), and from sulphocyanic acid by the circumstance that the red solution of sulphocyanide of iron, on the addition of ferridcyanide of potassium, and on being warmed, very soon precipitates Prussian blue, which is not the case with any other persalt of iron.

### *Physiological Relations.*

*Occurrence.*—We learn from pure chemistry that acetic acid is formed in various processes of decomposition of vegetable substances—in their fermentation as well as in their dry distillation: we shall, however, presently see that it often occurs as a product of distillation of several nitrogenous animal substances. It was formerly believed that it much more frequently existed pre-formed in the animal juices than has now been shown to be the case. On this point there was formerly a controversy between Gmelin and Berzelius; the former regarding the acid which formed the soluble salts occurring in the animal fluids as acetic acid, while the latter maintained it was lactic acid; Gmelin's idea was that the volatility of the acetic acid was heightened by its combination with an organic matter. The question has finally been settled in favor of the view maintained by Berzelius.

I have never been able to recognize it as a normal constituent in any of the animal juices. Scherer has however found it, as I have already mentioned (p. 57), in the juice of flesh, together with other acids of this group. It may often occur in the gastric juice in cases of disordered digestion. In a case where, after vegetables and a little meat, but no vinegar had been taken, the vomited matters were analyzed, and I satisfied myself with certainty regarding the presence of acetic acid. It has often been observed by others in vomited matters, but its presence has not always been demonstrated with sufficient chemical accuracy; for, on the one hand, vinegar or brandy might have been taken previously to the vomiting, or on the other hand, this acid might be confounded with metacetic or butyric acid. The proof that spirit of wine is converted in the stomach into acetic acid during normal digestion, will be given when we treat of the process of gastric digestion.

Bouchardat and Sandras<sup>1</sup> think that they have sometimes discovered traces of acetic acid in the blood of animals whose food has been steeped in brandy.

The answer to the question, what change acetic acid undergoes in the animal organism when conveyed into it from without, belongs to the department of pure physiological chemistry.

Whether the acids of this group found by Scherer in the fluids of flesh have their origin in the fleshy fibre which has become effete, or whether they arise from the decomposition of other matters, and are only isolated in the muscular juice, are questions which can only be decided by further investigation.

### METACETONIC ACID.— $C_6H_5O_3.HO$ .

#### *Chemical Relations.*

*Properties.*—This acid, which has also been named *butyro-acetic acid* and *propionic acid*, forms, when in a concentrated state, a colorless, oily fluid, which at a low temperature solidifies in a crystalline form, boils at about  $140^\circ$ , has a peculiar sauer-kROUT-like taste, and in its general character deports itself like the acids of this group; it is not perfectly soluble in a small quantity of water, but forms oily drops on it.

*Composition.*—According to the above formula it consists of:

Carbon, . . . . .	6 atoms,	. . . . .	48.649
Hydrogen, . . . . .	5 "	. . . . .	6.757
Oxygen, . . . . .	3 "	. . . . .	32.432
Water, . . . . .	1 "	. . . . .	12.162
			100.000

The atomic weight of the hypothetical anhydrous acid = 815.5; its saturating capacity = 12.31.

According to the investigations of Kolbe, to which we have already referred, this acid may, or indeed must be regarded as ethyloxalic acid =  $C_4H_5.C_2O_3.HO$ .

*Combinations.*—With bases this acid forms soluble salts of a fatty and glistening appearance, some of them also conveying a fatty feeling to the touch.

*Metacetate of baryta* crystallizes in small rectangular octohedra or rectangular prisms with oblique terminal surfaces.

*Metacetate of silver* forms glistening white granules or small prisms, which are little changed by the action of light, are difficult of solution in water, and when heated fuse, and at length noiselessly smoulder away.

Metacetate of oxide of ethyl in contact with ammonia becomes converted into the colorless crystalline substance called *metacetamide*,  $H_2N.C_6H_5O_2$ , which, by the agency of anhydrous phosphoric acid, is converted, more easily even than metacetate of ammonia, into cyanide of ethyl.

<sup>1</sup> Ann. de Chem. et de Phys. 3 Sér., T. 21, pp. 448-457.

*Metacetone*,  $C_6H_8O$ , cannot be obtained from metacetic acid, but is yielded by the decomposition of one part of sugar or starch with three parts of caustic lime; it forms a colorless, oily, volatile fluid that is essentially different from oxide of ænyl which is isomeric with it.

*Aldehyde of metacetic acid*,  $C_6H_8O.HO$ , was discovered by Guckelberger,<sup>1</sup> among the products of distillation, during the oxidation of nitrogenous matters by sulphuric acid and peroxide of manganese; it is a colorless fluid, having an ethereal odor; its specific gravity = 0.79, it boils at about 50°, is miscible with water in every proportion, gradually becomes acid when exposed to the air, but does not reduce a solution of a silver-salt; hence, it is still questionable whether this fluid should be ranked among the aldehydes.

*Preparation.*—Metacetic acid is formed during the spontaneous decomposition of many vegetable substances, as for instance, peas, lentils, and tan; by the action of hydrated potash on sugar, starch, gum, &c.; also during the fermentation of tartrate of lime in contact with nitrogenous bodies, in the decomposition of cyanide of ethyl by caustic potash; and lastly (and, in a zoo-chemical view, this mode of its formation is the most important), in the oxidation of fats by nitric acid (Redtenbacher),<sup>2</sup> in the oxidation of albuminous bodies by chromic acid, or by sulphuric acid and peroxide of manganese (Guckelberger),<sup>3</sup> and in the fermentation of glycerin, the well-known product of decomposition of the fats, by means of common yeast (Redtenbacher).<sup>4</sup> This acid is obtained most easily and in the purest form either by distillation of the product of the fermentation of yeast and glycerin, or by treating metacetone with chromic acid or hydrated potash; otherwise, it is ordinarily prepared by treating 1 part of sugar with 3 of hydrated potash, in which, however, it has to be separated from the other acids which are simultaneously developed, namely oxalic, formic, and acetic acids.

*Tests.*—Metacetic acid must, in the first place, be separated by distillation from other non-volatile organic substances with which it may have been mixed, and then by oxide of mercury, from any formic acid that may be present. If acetic acid be also present, the best method is to combine both acids with soda, when, on evaporating the saline solution, the acetate crystallizes sooner than the metacetate. The salt which metacetic acid forms with lead is not crystallizable, while, as every one knows, the acetate of lead crystallizes very readily. How this acid is to be separated and distinguished from the remaining acids of this group, will be described when we treat of those acids. Since, however, nothing can be concluded regarding the identity of any given substance with metacetic acid either from the forms of its salts, which have not yet been determined with crystallographic accuracy, or from the boiling-point of the fluid, it is only by the elementary analysis of a pure salt that the presence of metacetic acid can be scientifically determined.

As we proceed in the subject of zoo-chemistry, we shall become acquainted with a number of bodies whose characteristic properties are so

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 64, S. 46 ff.

<sup>2</sup> Ibid. Bd. 64, S. 46 ff.

<sup>3</sup> Ibid. Bd. 59, S. 41–57.

<sup>4</sup> Ibid. Bd. 57, S. 174–177.



The atomic weight of the hypothetical anhydrous acid = 987.5; its saturating capacity = 10.126.

According to the beautiful investigations of Kolbe, butyric acid may be regarded as an oxalic acid conjugated with the carbo-hydrogen  $C_6H_7 = C_6H_7 \cdot C_2O_3 \cdot HO$ .

*Combinations.*—The alkaline butyrates are deliquescent, and not crystallizable; the compounds of butyric acid with the metallic oxides lose a portion of their acid when heated, and even at an ordinary temperature evolve a strong odor.

*Butyrate of baryta*,  $BaO \cdot \overline{Bu} + 4HO$ , crystallizes in smooth prisms, grouped together in a wart-like form, and having a fatty glistening appearance; it retains its water of crystallization at  $100^\circ$ , and dissolves readily in water; if thrown in small pieces on water, it assumes, like camphor, a rotatory motion till it is dissolved; further, it turns red litmus blue.

*Butyrate of lime*,  $CaO \cdot \overline{Bu} + HO$ , crystallizes in fine needles; it has the odor of butyric acid, dissolves readily in cold water, but separates almost entirely on boiling, and on dry distillation yields bodies similar to ethereal oils, namely, *butyrone*,  $C_7H_7O$ , and *butyral*,  $C_8H_8O_2$ .

*Butyrate of magnesia*,  $MgO \cdot \overline{Bu} + 5HO$ , forms white plates resembling boracic acid.

*Butyrate of zinc* decomposes on boiling into a strongly basic insoluble salt.

*Butyrate of copper*,  $CuO \cdot \overline{Bu} + 2HO$ , occurs in eight-sided, bluish-green prisms, has a strong odor of butyric acid, and is only slightly soluble in water. At a temperature of about  $100^\circ$  most of the acid is expelled from this salt.

*Butyrate of lead* does not crystallize, and is only to be obtained in a syrup form.

*Butyrate of silver* forms white nacreous plates, is almost insoluble, and smoulders at a glow-heat without explosion.

*Butyramide*,  $H_2N \cdot C_8H_7O_2$ , is obtained from butyrate of oxide of ethyl when acted on by ammonia; it forms colorless crystalline tablets, which resist the action of the atmosphere; it communicates a taste which is at first sweetish but afterwards bitter; it fuses at  $115^\circ$ , and at a higher temperature sublimes without change; it is soluble in water, alcohol, and ether; by anhydrous phosphoric acid it is converted into *butyronitrile*,  $C_8H_7N$ , whose theoretical formula, according to Kolbe, must =  $C_6H_7 \cdot C_2N$ . Butyronitrile is an oily fluid, with an agreeable, somewhat aromatic odor; its specific gravity is 0.795, and its boiling point  $118.5^\circ$ ; treated with potassium it yields cyanide of potassium, hydrogen, and certain carbo-hydrogens.

*Aldehyde of butyric acid*,  $C_8H_7O \cdot HO$ , has hitherto only been found by Guckelberger,<sup>1</sup> in the products which are obtained by the action of peroxide of manganese and sulphuric acid on albuminous or gelatinous substances. It is a colorless fluid, its specific gravity is 0.8, and its boiling-point  $68^\circ$ ; it is slightly soluble in water, but dissolves freely in

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 64, S. 46 ff.



alcohol and ether; it soon becomes acid when exposed to the air; it reduces solutions of the silver-salts, and, like aldehyde of acetic acid, it yields with ammonia a crystallizable compound,  $\text{H}_3\text{N} \cdot \text{C}_4\text{H}_7\text{O} \cdot \text{HO} + 10 \text{ aq.}$

*Butyrate of glycerin* has been prepared by Pelouze and Gélis,<sup>1</sup> by gently heating butyric acid and glycerin with concentrated sulphuric acid, and separating the new compound from the mixture by means of water; or by passing hydrochloric acid gas through a mixture of butyric acid and glycerin; on the addition of water it separates as a yellow oil, soluble in concentrated alcohol and ether, which, when treated with caustic alkalis, again resolves itself into butyric acid and glycerin. Whether this body be identical with the butyrin (butyrate of oxide of lipyl) occurring in the fat of milk but not yet isolated, cannot at present be decided, since no elementary analysis of it has been instituted.

*Preparation.*—Butyric acid, which was originally discovered by Chevreul in the products of the saponification of butter, is also formed when this substance becomes rancid, and occurs amongst the products of decomposition when oleic acid is submitted to dry distillation, and especially when it is acted on by fuming nitric acid; it is likewise produced from non-fatty nitrogenous matters, as albumen, fibrin, and gelatin, during their putrefaction or their decomposition by strong oxidizing agents; and, contrary to expectation, it has been found in certain processes of fermentation of non-nitrogenous bodies, as starch and sugar, where the nitrogenous admixtures only act as ferments. Lactate of lime, in the presence of nitrogenous matter, becomes converted into butyrate of lime. To obtain pure butyric acid on a large scale, we should have recourse to the last-named method. The most simple mode of procedure is to expose carob (the fruit of *Ceratonium siliqua*), or sugar, with sour milk and a little cheese, and with some carbonate of lime, at a temperature of  $30^\circ$  to  $35^\circ$ , as long as gas continues to be evolved, namely for five or six weeks; the filtered fluid is then decomposed with carbonate of soda, which causes a precipitation of carbonate of lime; the solution of butyrate of soda is now strongly concentrated, and, after being decomposed with sulphuric acid, is distilled; finally, the butyric acid is freed from water and acetic acid by fused chloride of calcium.

*Tests.*—This acid must first be separated by distillation from the non-volatile substances, as, for instance, lactic acid, with which it is not unfrequently associated; in the distillate we can then only have the acids of this group. We shall here refer to the means of distinguishing it from the acids which have been already described, namely, formic acid, acetic acid, and metacetic acid. The first may be very easily removed by means of its property (to which we have frequently referred) of reducing the oxides of the noble metals. The acids must then be combined with soda, when the greater part of the acetate of soda may be removed by crystallization. The soda-salts of the mother-liquid are afterwards to be decomposed by tolerably concentrated sulphuric acid, yielding in the receiver metacetic and butyric acids, with a little acetic acid; from these the butyric acid may be pretty well separated by fractional distillation, since that which passes over at  $140^\circ$  is only metacetic acid,

<sup>1</sup> L'Institut. No. 494.

with traces of acetic acid, and it is not till the temperature is raised to  $160^{\circ}$  or  $165^{\circ}$ , that tolerably pure butyric acid enters the receiver. If other analogous acids be also present, we must not be contented with this mode of procedure; specific as it may appear to be, we must not rely on the peculiar odor of butyric acid, but we must convert the butyric acid into one of the above-described butyrates, and after comparing the salt thus obtained with the corresponding salt of pure butyric acid, we must institute an elementary analysis, or at the least we must determine the atomic weight or the saturating capacity.

The atomic weight of the hypothetical anhydrous butyric acid is 987.5 (for 8 at. carbon=600.0, 7 at. hydrogen=87.5, and 3 at. oxygen=300). Now if, in a baryta-salt, we have found 49% of baryta and 51% of butyric acid, then 49 : 51 must be the ratio in which the known atomic weight of baryta (=955.3) stands to the atomic weight of butyric acid ( $49 : 51 :: 955.3 : x$ )=994.4.

By a similar determination of the quantity of a base contained in a salt, we calculated the saturating capacity, by which, as is well known, we understand the number which expresses the quantity of oxygen contained in that quantity of base which is required by 100 parts of an anhydrous acid to form a neutral salt. Hence the saturating capacity of butyric acid is=10.126. If we regard the above instance as an empirical result, 49 BaO saturate 51 Bu, or 100 Bu saturate 96.076 BaO; in this, however, there are contained 10.06 parts of oxygen, which is a tolerably close approximation to the required number.

### *Physiological Relations.*

*Occurrence.*—In the contents of the stomach, or rather in food which has been ejected by vomiting, we sometimes meet with a nauseous acrid or rancid-smelling volatile acid, which, beyond all question, is butyric acid. Tiedemann and Gmelin often obtained a fluid resembling butyric acid, by distillation of the contents of the stomachs of sheep, oxen, and horses, fed with oats. Since the contents of the stomach can pass into the acetous, and, as we shall presently see, also into the lactic fermentation, there is nothing surprising in the circumstance of their also passing into the butyric fermentation; but, even in abnormal conditions, butyric acid has not been recognized in the contents of the stomach with that absolute certainty, which is as necessary in physiologico-chemical researches as in all other departments of natural inquiry.

Free butyric acid was long ago discovered in the urine by Berzelius, who, however, did not think that it was often to be found there. In the urine of pregnant women, and of those who, after delivery, do not suckle their children, I have sometimes found butyric acid; or, at all events, a fat which, on saponification, yielded a volatile acid, with the odor of butyric acid.

In the sweat, especially in that of the genitals and lower extremities of corpulent persons, we find volatile matters, with an acid reaction, and having an odor partly of butyric acid and partly of other acids of this group. Berzelius thought that the acid reaction was due to butyric acid alone; but, in the present state of our knowledge, it must remain

doubtful whether the homologous, highly carbonaceous acids do not occur in the sweat, with or in place of butyric acid. In examining the watery extract of a night-dress steeped in perspiration, taken from a woman a few days after delivery, I found, on saponification, a rancid-smelling, volatile acid.

Schottin has determined the existence of butyric acid in the sweat with all the necessary accuracy, his investigations having been carried on under my own superintendence. Its quantity was, however, far less than that of the acetic and formic acids. It is, moreover, not a mere product of decomposition of the secretion of the sebaceous follicles, as I formerly believed, but occurs in a free state in the sweat of the axillary regions, the genitals, and the feet.

In the *milk*, in addition to other fats, as olein and margarin, there occurs a fat which has never yet been isolated in a state of purity, and which, on saponification, yields butyric acid, together with other acids of this group, namely, caproic, caprylic, and capric acids. The best investigations in reference to this substance were made, first by Chevreul, in his classical work on the fats; subsequently by Bromeis;<sup>2</sup> and lastly by Lerch,<sup>3</sup> under the direction of Redtenbacher. Even in butter there is only a little of this substance, which yields butyric acid. From 100 parts of tolerably pure butyrin, Chevreul<sup>4</sup> only obtained 7 parts of volatile acids; Simon<sup>5</sup> and Herberger<sup>6</sup> were able to obtain only very minute quantities of volatile acids from the fat of woman's milk.

That there are fats in the *blood* which, on saponification, yield volatile acids, may be demonstrated by any one who operates with care on large quantities of the fatty matter collected from this fluid. From the blood taken from a woman within the first few days after her delivery, I obtained, by distillation with dilute sulphuric acid, volatile acids whose general properties coincided with those of this group.

[Free butyric acid has likewise been detected in the *faeces* by Ragsky and Percy.<sup>7</sup>—G. E. D.]

*Origin.*—After what has been stated regarding the different ways in which butyric acid may be formed, we need not wonder that it is sometimes met with in the *primæ viæ*; since it may, and indeed must principally be formed from the non-nitrogenous constituents of the food. The belief that farinaceous and saccharine foods are converted into butyric acid in the *primæ viæ*, and that they thus constitute the first step in the formation of fat, is based on a fiction regarding the possible formation of fat in general, which is at present devoid of any scientific proof. No one has as yet succeeded in ascertaining the presence of butyric acid, either in the *primæ viæ* or in the chyle; we know not what becomes of the other elements which are eliminated during the conversion of starch into butyric acid; and finally, chemically considered, butyric acid has no greater claim to the name of a fatty acid, than acetic or formic acid. We do not think that the conclusion can be justly deduced, that starch must be converted into butyric acid in order

<sup>1</sup> Recherches sur les corps gras.

<sup>2</sup> Ibid. Bd. 49, S. 212 ff.

<sup>3</sup> Frauenmilch, S. 41.

<sup>7</sup> Chemical Gazette. Vol. 8, p. 104.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 42, S. 46 ff.

<sup>4</sup> Recherches sur les corps gras, p. 193.

<sup>6</sup> Brande's Arch. Bd. 20, S. 3.

to be transformed into fat, simply because it accidentally happens that butyric acid was first prepared from a (very rarely occurring) fat, for we know that it may just as easily be obtained from albuminous bodies, and in far larger quantities from gelatin.

There is much stronger evidence in favor of the view which regards the butyric acid found in the blood, sweat, and urine, as a product of decomposition, arising from the disintegration of nitrogenous animal matters, effected by the oxygen dissolved in the juices (in the same way as the acid is formed from these substances by artificial means), or as probably resulting from a gradual oxidation of some of the carbo-hydrogens of the fats. This latter view is, however, only an hypothesis; but it is supported by the simplest induction. The fats are almost all combinations of fatty acids with a haloid base, glycerin or oxide of lipyl; these acids are, however, so similarly constituted to those of this group, that they have the same general formula  $=C_nH_{n-1}O_3.HO$ , with only this difference, that the carbo-hydrogens pertaining to them are expressed by higher atomic numbers (thus, for instance, margaric acid  $=C_{34}H_{33}O_3.HO$ ). In the complicated apparatus of oxidation which we recognize in the animal organism, the fats do not burn like the oil in the wick of a lamp, but they undergo an extremely gradual oxidation, as we learn from direct experiments, which have given us a knowledge of a very large number of fatty acids, with the most varied polymeric carbo-hydrogens, or, if we please so to express it, in the lowest stages of oxidation. From experiments instituted on this group of acids, we may assume that in the gradual oxidation,  $C_2H_2$  is always abstracted from the radical of margaric acid, and that this gradual abstraction may proceed with various degrees of rapidity, so that, in our investigations, we meet with carbo-hydrogen compounds of a lower order, which then progressively pass into the carbo-hydrogens of the acids of this group. As the radical  $C_4H_5$  of ethyloxalic acid passes into methyloxalic acid, we are justified in believing that the radical of margaric acid passes into cetylic acid. A gradual decarbonization of the fats must occur in the animal organism; and there are at present no scientific reasons for assuming that it takes place in any other way than that which has been described. We regard butyric acid, and the acids analogous to it, in so far as they occur in the animal body, as products of regressive metamorphosis of tissue, while in the different fatty acids of the vegetable kingdom the progression gradually ascends, step by step, to margaric acid.

### VALERIANIC ACID.— $C_{10}H_9O_3.HO$ .

#### *Chemical Relations.*

*Properties.*—This acid possesses the general properties of this group, has a well-known characteristic odor, an acrid burning taste, and produces a white spot upon the tongue; it does not become solid at a temperature of  $-15^\circ$ ; it boils at  $176^\circ$ , and dissolves in 26 parts of water: it also forms a second hydrate  $=Va.3HO$ .

*Composition.*—According to the above formula it consists of:

Carbon,	.	.	.	.	10 atoms,	.	.	.	.	58.824
Hydrogen,	.	.	.	.	9 "	.	.	.	.	8.823
Oxygen,	.	.	.	.	3 "	.	.	.	.	23.530
Water,	.	.	.	.	1 "	.	.	.	.	8.823
										<hr/> 100.000

The atomic weight of the hypothetical anhydrous acid=1162.5; its saturating capacity=8.602. According to Kolbe's hypothesis, its theoretical formula= $C_8H_9.C_2O_3.HO$ .

*Combinations.*—The valerianates are for the most part soluble: the alkaline salts do not crystallize, but most of the other salts crystallize in nacreous plates, similar to cholesterin or boracic acid; they have a sweetish, but at the same time a valerian-like taste. Valerianic acid is separated from its salts by acetic and succinic acids, but not by benzoic acid. The *lime-salt* effloresces on exposure to the air; the *zinc-salt* dissolves in 160 parts of water, and in 60 parts of spirit of wine; the aqueous solution becomes turbid when warmed, but clears again upon cooling; moreover it reddens litmus. The *silver-salt* is very insoluble.

*Valeronitrile*,  $C_{10}H_9N$  (or  $C_8H_9.C_2N$ ), was first discovered by Schlieper,<sup>1</sup> in the oxidation of gelatin by chromic acid; it may, however, be obtained from valerianate of ammonia, or valeramide ( $H_2N.C_{10}H_9O_2$ ), by anhydrous phosphoric acid. It is a thin, liquid, colorless, strongly refracting oil, smelling like alder leaves, and having a hot aromatic taste; its specific gravity is=0.81; it boils at 125°, inflames readily, dissolves in water, alcohol, and ether, and, when treated with potassium, yields cyanide of potassium, hydrogen, and carbo-hydrogens.

*Valeral*,  $C_{10}H_{10}O_2$ , is produced by the dry distillation of valerianate of baryta; it is a very fluid inflammable oil, which, on exposure to the air, soon becomes converted into valerianic acid.

*Preparation.*—This acid occurs preformed in certain plants; it is, however, like the preceding acids, a not unfrequent product of decomposition both of vegetable and animal substances: it is obtained from fusel-oil (hydrated oxide of amyl) in precisely the same manner as acetic acid is obtained from alcohol (hydrated oxide of ethyl), and from oil of valerian by simple oxidation by means of an alkali; it is formed, together with other acids of this group, from the fats by oxidizing them with fuming nitric acid (Redtenbacher);<sup>2</sup> from animal nitrogenous matters, both by putrefaction (Iljenko and Laskowski),<sup>3</sup> and on decomposing them by strong oxidizing agents (Schlieper,<sup>4</sup> Guckelberger,<sup>5</sup> Liebig);<sup>6</sup> and finally, if leucine be treated with caustic potash, or allowed to putrefy, it becomes converted into valerianic and no other acid, ammonia and hydrogen being evolved.

It is most easily obtained in a state of purity by the action of spongy platinum and atmospheric air on potato fusel-oil.

*Tests.*—In most of the ways in which valerianic acid is formed, it occurs mixed with other acids of this group; and it is as impossible in

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 59, S. 1-32.

<sup>2</sup> Ibid. Bd. 55, S. 78-95, and Bd. 63, S. 264-278.

<sup>3</sup> Ibid. Bd. 64, S. 50.

<sup>4</sup> Ibid. Bd. 59, S. 41-57.

<sup>5</sup> Ibid. Bd. 59, S. 375-378.

<sup>6</sup> Ibid. Bd. 57, S. 127-129.

this case, as in that of the homologous acids, to detect it in a mixture by any special reagent; it must, therefore, be separated from these acids before it can be accurately examined. As its boiling-point is so high, it can readily be separated from the first-described acids of this group by fractional distillation; it may still remain contaminated with butyric acid, from which it can be tolerably well separated by crystallization of the baryta-salts, the valerianate and butyrate of baryta assuming different forms. But an elementary analysis, or a determination of the atomic weight must be made with the valerianate thus obtained, since mistakes may very easily arise between the salts of valerianic acid and those of certain acids afterwards to be described.

[Liebig<sup>1</sup> has recently published a paper on the separation of valerianic, acetic, and butyric acids, to which we may refer the reader.—G. E. D.]

### *Physiological Relations.*

*Occurrence.*—Although this acid is so easily and so variously obtained from animal substances, it has never yet been found preformed in the animal organism; and it is a striking fact that, so far as we yet know, the acids of this group, whose amount of carbon is divisible only by 2, and not by 4, are not found in the animal organism.

We shall consequently only have occasion to refer to these acids in the following pages, inasmuch as they sometimes occur as products of the artificial decomposition of animal substances.

### • CAPROIC ACID.— $C_{12}H_{22}O_2$ .HO.

*Properties.*—It is a somewhat thin liquid, with an odor resembling sweat; its specific gravity at  $+26=0.922$ ; it remains fluid at  $-9^\circ$ , boils at  $202^\circ$ , and dissolves somewhat difficultly in ether.

*Composition.*—According to its formula it consists of:

Carbon,	.	.	.	12 atoms,	.	.	.	62.069
Hydrogen,	.	.	.	11 "	.	.	.	9.483
Oxygen,	.	.	.	3 "	.	.	.	20.689
Water,	.	.	.	1 "	.	.	.	7.759
								<hr/> 100.000

The atomic weight of the anhydrous acid= $1337.5$ ; its saturating capacity= $7.476$ . According to the views of Kolbe, this acid should hypothetically be regarded as amyloxalic acid= $C_{10}H_{11}.C_2H_3.HO$ .

*Combinations.*—The caproates have the same taste and smell as the acid itself; and are mostly soluble in water and crystallizable. The *baryta-salt* crystallizes in long silky needles, united in tufts, is anhydrous, and unaffected by exposure to the atmosphere; the *silver-salt* is not crystallizable, and is very difficult of solution.

*Preparation.*—Like butyric acid, this acid is not only formed when butter is saponified or becomes rancid, but also when oleic acid is decomposed by fuming nitric acid, and when albuminous bodies are acted on by peroxide of manganese or bichromate of potash and sulphuric acid. In the

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 71, S. 355.

products of the decomposition of saponified butter we find caproic acid mixed with butyric, caprylic, and capric acids, which may be removed by the crystallization of their baryta-salts. On boiling the dried mass of the baryta-salts with 5 or 6 parts of water, the butyrate and caproate are taken up, while the salts of caprylic and capric acid remain undissolved. The caproate of baryta is the first to crystallize from the solution, and the acid may easily be isolated from the salt.

*Tests.*—The caproate of baryta not only crystallizes sooner than the butyrate, but also sooner than the valerianate, if this should happen to be present; caproate of baryta forms small clusters, consisting of microscopic prisms, while the valerianate, as we have already mentioned, appears in minute plates like cholesterin. This separation of caproic acid from its allied acids, is more easily explained theoretically than effected practically. There are no special means of determining the presence of caproic acid, except by an elementary analysis, and the determination of the atomic weight.

### *Physiological Relations.*

*Occurrence.*—The remarks which we made regarding the occurrence of butyric acid in the animal organism, apply equally to caproic acid. From its peculiar sweat-like odor, it is not improbable that it exists in sweat; but of this we have as yet no proof. No one, so far as I know, has yet sought for it in the urine or in the contents of the stomach. In our observations on butyric acid we alluded to the fatty matters contained in the milk, and probably also in the blood, which, on saponification, yield this acid.

### ÆNANTHYLIC ACID.— $C_{14}H_{13}O_3 \cdot HO$ .

#### *Chemical Relations.*

*Properties.*—It is a colorless oily liquid, of a faint aromatic odor and taste; it boils at about  $215^{\circ}$ , may be distilled with only partial decomposition, dissolves slightly in water, and when inflamed burns with a clear but smoky flame.

*Composition.*—According to the above formula it consists of:

Carbon,	:	:	:	:	14 atoms,	:	:	:	:	64.615
Hydrogen,	:	:	:	:	13 "	:	:	:	:	10.000
Oxygen,	:	:	:	:	8 "	:	:	:	:	18.462
Water,	:	:	:	:	1 "	:	:	:	:	6.923

100 000

The atomic weight of the hypothetical anhydrous acid= $1512.5$ , and its saturating capacity= $6.611$ . Its rational formula= $C_{12}H_{13} \cdot C_2O_3 \cdot HO$ .

*Combinations.*—With the exception of the alkaline salts, most of its salts are difficult of solution, generally resembling tablets of cholesterin: moreover this acid has a strong tendency to form acid salts. The *baryta-salt* crystallizes in nacreous scales, which are soluble in water and in alcohol.

*Œnanthylous acid*,  $C_{14}H_{13}O_2.HO$ , formerly also named œnanthic acid, occurs combined with oxide of ethyl in various fusel oils, especially in that of wine. Whether it be actually to be regarded as a lower state of oxidation of œnanthylic acid, or as a special acid, cannot at present be decided.

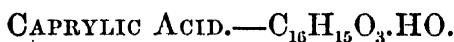
*Œnanthal*, *aldehyde of œnanthylic acid*,  $C_{14}H_{14}O_2$ , is obtained by the simple distillation of castor oil; like the other aldehydes, when exposed to the atmosphere, it readily oxidizes into the corresponding acid, and forms a compound (although somewhat unstable) with ammonia.

*Preparation*.—This acid, which Laurent formerly discovered amongst the products of distillation of the oils, and named *azoleic acid*, is formed, together with other acids of this group, during the decomposition of wax, oleic acid, and especially of castor oil, by concentrated nitric acid. In using castor oil, however, we obtain this acid unmixed with any others, so that we have only to combine it with baryta, and recrystallize the salt, in order to obtain it in a state of purity.

*Tests*.—As the baryta-salt of this acid separates from the mother-liquid earlier than caproate of baryta, and more slowly than the caprylate, and as, further, it crystallizes in plates, while the two latter salts form minute needles, which are grouped together so as to have a wart-like appearance, we have a means of separating, at least roughly, this acid from those which are most closely allied to it. We cannot, however, be perfectly certain regarding its actual presence, without an elementary analysis, or the determination of its atomic weight.

#### *Physiological Relations.*

*Occurrence*.—As has been already mentioned, this acid is only of interest in relation to animal physiology, inasmuch as it is one of the products of oxidation of the fats; and the observations which were made regarding the occurrence of valerianic acid are here equally applicable, except that œnanthylic acid is not produced during the decomposition of nitrogenous complex atoms.



#### *Chemical Relations.*

*Properties*.—At the ordinary temperature this acid forms a soft, semi-fluid mass, which crystallizes in needles below  $+10^{\circ}$ , boils at  $236^{\circ}$ , has a sweat-like odor, and acid and acrid taste, is difficult of solution in water, and is inflammable.

*Composition*.—According to the above formula it consists of:

Carbon,	.	.	.	.	.	16 atoms,	.	.	.	.	66.667
Hydrogen,	.	.	.	.	.	15 "	.	.	.	.	10.416
Oxygen,	.	.	.	.	.	8 "	.	.	.	.	19.667
Water,	.	.	.	.	.	1 "	.	.	.	.	6.250
											100.000

The atomic weight of the anhydrous acid=1687.5, and its saturating capacity=5.926. Its rational formula is  $C_{14}H_{15}.C_2O_3.HO$ .



*Combinations.*—The salts of this acid are more difficult of solution than the corresponding salts of the acids already described. Its *baryta-salt* crystallizes in white granules of the size of poppy-seeds, is anhydrous, resists the action of the atmosphere, and does not fuse at 100°. The *silver-salt* is white and almost insoluble. The *lead-salt* is also very difficult of solution.

*Caprylone*,  $C_{15}H_{31}O$ , was discovered by Guckelberger<sup>1</sup> among the products of the dry distillation of caprylate of baryta; it crystallizes in fine needles of a silky lustre, but when fused resembles Chinese wax; it is perfectly white, fuses at 40°, solidifies at 38°, and boils at 178°, is devoid of taste, has a waxy smell, is lighter than water and insoluble in it, but dissolves readily in strong alcohol, in ether, and in ethereal as well as fatty oils. With nitric acid of 1.4 specific gravity it yields an acid nitrogenous oil (*nitro-caprylonic acid*?)

*Preparation.*—We have become acquainted with this acid as a product of the saponification of butter, and as a product of the oxidation of oleic acid when acted on by nitric acid; as in the latter case it is mixed with several substances, it is best obtained by the recrystallization of the baryta-salts of the volatile acids of butter. In the observations on caproic acid it was mentioned that the dry mass of the baryta-salts of all four acids, when treated with five or six parts of water, separates into a soluble portion containing the butyrate and caproate, and an undissolved portion, containing the caprylate and caprate of baryta. If now the undissolved portion be dissolved in boiling water and filtered while still hot, most of the caprate separates while the caprylate remains in solution. In order to effect a perfect purification, the baryta-salt must be several times recrystallized before we separate the acid from it.

*Tests.*—We must separate the caprylic acid from the other acids in the manner just described, and then determine the atomic weight.

### *Physiological Relations.*

*Occurrence.*—All that has been remarked regarding the physiological relations of butyric and caproic acids applies equally to caprylic acid.

### PELARGONIC ACID.— $C_{18}H_{35}O_2$ .

#### *Chemical Relations.*

*Properties.*—It is an oily, colorless fluid, which at a lower temperature than +10° becomes solid, but liquefies at and above that temperature; it has a faint odor resembling that of butyric acid, is almost insoluble in water, but communicates to it an acid reaction, and boils at about 232°.

*Composition.*—In accordance with the above formula it consists of:

Carbon, . . . . .	18 atoms,	68.850
Hydrogen, . . . . .	17 "	10.760
Oxygen, . . . . .	8 "	15.190
Water, . . . . .	1 "	5.700

100 000

<sup>1</sup> Ann. d. Ch. u Pharm. Bd. 69, S. 201-6.

The atomic weight of the anhydrous acid = 1862.5; its saturating capacity = 5.369; its rational formula =  $C_{16}H_{17}.C_2O_3.HO$ .

*Combinations.*—The *baryta-salt* of this acid crystallizes like the valerianate and oenanthylate of baryta in glistening scales; it contains no water of crystallization, is unaffected by the atmosphere, and is less soluble than the oenanthylate and caprylate of baryta, but rather more soluble than the caprate.

*Preparation.*—As this acid, unmixed with other volatile acids, occurs in the leaves of *Pelargonium roseum*, its preparation from that plant is preferable to that from the products of decomposition of oleic and choloidic acids by nitric acid, amongst which it was first discovered by Redtenbacher.<sup>1</sup> Gerhardt<sup>2</sup> has obtained this acid by oxidizing oil of rue,  $C_{20}H_{19}O_3$ , with nitric acid.

*Tests.*—By the crystallization of its baryta-salt we must prepare this acid so that we can make an elementary analysis and determine its atomic weight.

#### *Physiological Relations.*

The remarks already made regarding the physiological relations of oenanthylic acid are equally applicable here.

#### CAPRIC ACID.— $C_{20}H_{19}O_3.HO$ .

##### *Chemical Relations.*

*Properties.*—Little is yet known regarding this acid in a state of purity, for what was formerly regarded as capric acid was a mixture of capric and caprylic acids. It constitutes a soft, greasy mass which fuses at  $+30^\circ$ , and evolves a faint goat-like odor, is somewhat soluble in hot water, but separates on cooling in glistening crystalline particles; its boiling-point is higher than that of any of the other acids of this group, but is considerably below  $300^\circ$ .

*Composition.*—According to the above formula it consists of:

Carbon, . . . . .	20 atoms,	. . . . .	69.767
Hydrogen, . . . . .	19 "	. . . . .	11.046
Oxygen, . . . . .	3 "	. . . . .	13.954
Water, . . . . .	1 "	. . . . .	5.233

100.000

The atomic weight of the hypothetical dry acid = 2037.5; its saturating capacity = 4.909; its rational formula =  $C_{18}H_{19}.C_2O_3.HO$ .

*Combinations.*—The salts of this acid are more insoluble than those of the other acids of this group. The *baryta-salt* crystallizes in delicate, glistening needles; it is unaffected by exposure to the atmosphere, and contains no water.

Oil of rue,  $C_{20}H_{19}O$ , the ethereal oil of *Ruta graveolens*, may be regarded as anhydrous aldehyde of capric acid; in point of fact it is con-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 59, S. 41–57, and Bd. 57, S. 170–174.

<sup>2</sup> Ann. de Ch. et de Phys. T. 24, pp. 112–116.

verted into capric acid by the action of nitric acid; but by more prolonged action, into pelargonic acid.

*Preparation.*—This may be readily inferred from what has been stated regarding the preparation of caprylic acid.

*Tests.*—We must obtain a pure salt according to the method described in our observations on caprylic acid, and then analyze it. R. Wagner<sup>1</sup> has, however, discovered a method of detecting this acid when mixed with other substances; for on heating such a mixture with concentrated sulphuric acid, it always appears associated with its aldehyde, and on supersaturation with potash, an intense odor of oil of rue is developed.

Wagner has in this way discovered this aldehyde in butter, in cod-liver oil and other fish-oils, in old cheese, in a piece of herring, &c.

### *Physiological Relations.*

The remarks on the physiological relations of caprylic acid apply equally to this acid.

In the saponification of butter we sometimes obtain only a single acid, *vaccic acid*,  $C_{20}H_{38}O_5 \cdot 2HO$  instead of butyric and caproic acids. This acid reduces silver-salts, and taking up 1 atom of oxygen, becomes converted into butyric and caproic acids ( $C_{20}H_{38}O_5 + O = C_8H_{14}O_3 + C_{12}H_{22}O_3$ ); it undergoes the same conversion when exposed to the atmosphere, and so also does its baryta-salt.

*Delphic and hircic acids* which were formerly regarded as independent acids are probably identical with, or mixtures of some of the acids of this group.

### CETYLIC ACID.— $C_{32}H_{64}O_3 \cdot HO$ .

#### *Chemical Relations.*

*Properties.*—The body, which is also known as *ethalic acid*, forms colorless glistening needles, fuses at  $57^\circ$ , but is solid at  $55^\circ$ , may be distilled without undergoing decomposition, and is insoluble in water.

*Composition.*—This acid, which is isomeric with the non-volatile *palmitic acid*, obtained from palm-oil, consists according to the above formula of:

Carbon, . . . . .	82 atoms,	75.000
Hydrogen, . . . . .	31 "	12.109
Oxygen, . . . . .	3 "	9.375
Water, . . . . .	1 "	3.516
		<hr/>
		100.000

The atomic weight of the hypothetical anhydrous acid = 3087.5; its saturating capacity = 3.239. This acid, which was originally discovered by Dumas and Stass,<sup>2</sup> has subsequently been accurately examined by Smith.<sup>3</sup>

If Kolbe's theory be applicable to this acid, cetylic acid must be re-

<sup>1</sup> Journ. f. pr. Ch. Bd. 46, S. 155-157.

<sup>2</sup> Ann. de Chim. et de Phys. T. 72, pp. 5-11.

<sup>3</sup> Ann. d. Ch. u Pharm. Bd. 42, S. 40-51.

garded as  $C_{30}H_{61}O_2.HO$ , which would explain why it differs from the isomeric palmitic acid. Two isomeric acids cannot appropriately be placed in the same group; hence we place cetylic acid here instead of considering it with the solid fatty acids. We also find in this relation an additional reason why the solid fatty acids whose general formula may be regarded as  $=C_nH_{n-1}O_2.HO$ , should not be regarded as simple continuations or ascending members of this group.

*Combinations.*—The alkaline salts of this acid are soluble in water, and crystallize readily in white nacreous scales.

*Preparation.*—Spermaceti, from which this acid is obtained, is a haloid salt like the other fats, but instead of this acid being combined with oxide of lipyl, it is united to another haloid base entirely corresponding with the ethers of pure chemistry; this haloid base when treated with solid caustic alkalies is converted into cetylic acid. We obtain the acid which exists preformed in the spermaceti, by saponifying the latter with a caustic alkali, decomposing the soap with hydrochloric acid and digesting the newly formed mixture of cetylic acid and ethal ( $C_{32}H_{65}O.HO$ ) with milk of lime; the ethal is then extracted with cold alcohol while the cetylate of lime remains. The lime-salt is then decomposed by hydrochloric acid, and the separated cetylic acid purified by solution in ether.

This haloid base, *ethal* or *hydrated oxide of cetyl*, which is obtained on the saponification of spermaceti, bears exactly the same relation to cetylic acid that alcohol bears to acetic acid or fusel oil to valerianic acid. Moreover, as we shall further more fully describe, cetylic acid may in a similar way be prepared from this body by heating one part of it in six parts of a previously heated mixture of equal parts of hydrated potash and caustic lime to a temperature of  $210^{\circ}$ – $220^{\circ}$ ; in this process, hydrogen is developed and an alkaline cetylate formed ( $C_{32}H_{65}O.HO + KO + HO = 4H + KO.C_{32}H_{61}O_2$ ) which must be purified by solution in water and crystallization, and then combined with baryta, from which on the addition of hydrochloric acid we can separate the cetylic acid.

*Tests.*—When the acid occurs pure and isolated, it is not difficult to distinguish it from other acids; its crystallizability and its comparatively high boiling-point distinguish it from the other acids of this group, and its volatility from the solid fatty acids. On finding it in a body in which it has not been previously recognized, we should always institute an elementary analysis, and determine its saturating capacity, since it is not only possible but very probable that several similar acids remain to be discovered.

#### *Physiological Relations.*

*Occurrence.*—This acid has hitherto only been found in an animal fat, namely spermaceti, in combination with hydrated oxide of cetyl; and in Japanese wax (Meyer), in combination with oxide of lipyl.

*Origin.*—If margaric acid were actually an acid homologous to cetylic acid and to the acids of this group generally, we might easily understand that cetylic acid was produced from this acid in the same manner as acetic is formed from metacetic acid; for margaric acid stands in the same relation to cetylic acid as metacetic acid does to acetic acid, the difference between each pair being  $C_2H_2$ .

It is impossible at present to form any conjectures regarding the special importance of these acids in the few positions in which they are principally deposited. For a description of *hydrated oxide of cetyl* see "*haloid bases and fats*."

### THE SUCCINIC ACID GROUP.



The acids of this group are only interesting in reference to zoo-chemistry, inasmuch as, like many acids of the previous group, they are products of decomposition of very common animal matters, and especially of fats. These acids may also be regarded as conjugated oxalic acids, combined with a carbo-hydrogen isomeric with olefiant gas; at least some of the reasons which have been advanced by Kolbe in support of the theoretical composition of the preceding group, favor this hypothesis. These acids, with their empirical and hypothetical formulæ, are as follows:

Succinic acid, . . . . .	$=C_4H_2O_3.HO=C_2H_2.C_2O_3.HO$
Lipic or pyrotartaric acid, . . . . .	$=C_5H_3O_3.HO=C_3H_3.C_2O_3.HO$
Adipic acid, . . . . .	$=C_6H_4O_3.HO=C_4H_4.C_2O_3.HO$
Pimelic acid, . . . . .	$=C_7H_5O_3.HO=C_5H_5.C_2O_3.HO$
Suberic acid, . . . . .	$=C_8H_6O_3.HO=C_6H_6.C_2O_3.HO$
Sebacic acid, . . . . .	$=C_{10}H_8O_3.HO=C_8H_8.C_2O_3.HO$

It is, moreover, worthy of remark, that the acids of this group, which contain an even number of atoms of carbon, form a series very analogous to the acids of the preceding group; the acid of one series differing from the corresponding acid of the other merely by one equivalent of hydrogen.

Succinic acid, . . . . .	$C_4H_2O_3+H=$	acetic acid, . . . . .	$C_2H_3O_2$
Adipic acid, . . . . .	$C_6H_4O_3+H=$	metacetic acid, . . . . .	$C_4H_5O_2$
Suberic acid, . . . . .	$C_8H_6O_3+H=$	butyric acid, . . . . .	$C_6H_7O_2$
Sebacic acid, . . . . .	$C_{10}H_8O_3+H=$	valeric acid, . . . . .	$C_8H_9O_2$

Moreover, the acids of this group (like those of the preceding group) are formed when oleic acid is oxidized by nitric acid.

These acids possess the following characters in common: they crystallize readily and well, do not fuse till they attain a temperature of from  $100^\circ$  to  $200^\circ$ , and at a higher temperature they sublime in needles, developing at the same time a suffocating vapor; moreover at an ordinary temperature they are devoid of odor, have an acid taste, dissolve readily in water, alcohol, and ether, and have an acid reaction,—none of them, with the exception of sebacic acid, are decomposed by boiling nitric acid: fused with hydrated potash, they yield oxalic acid together with volatile products. As in the preceding group, the solubility of their salts stands nearly in an inverse ratio to the height of the atomic weight of the acid. \*

As these acids are only of importance in animal chemistry as products of decomposition, and belong strictly to pure chemistry, we shall restrict

ourselves to the consideration of two of the most important of them, namely, *succinic* and *sebacic acids*. As, however, none of them occur preformed in the animal body, there is obviously nothing to be said regarding their physiological relations.<sup>1</sup>

### SUCCINIC ACID.— $C_4H_2O_3.HO$ .

*Properties*.—When perfectly anhydrous it occurs in very delicate needles, which fuse at  $145^\circ$  and boil at  $250^\circ$ ; with one atom of water (corresponding with the above formula), it crystallizes in oblique rectangular prisms which fuse at  $180^\circ$ , and sublime at  $250^\circ$  in the form of needles or plates, containing only half an atom of water, and fusing at  $160^\circ$ . In other respects, it has the common characters of this group.

*Composition*.—According to the above formula it consists of:

Carbon, . . . .	4 atoms,	. . . .	40.678
Hydrogen, . . . .	2 “	. . . .	3.890
Oxygen, . . . .	3 “	. . . .	40.678
Water, . . . .	1 “	. . . .	15.254

100.000

The atomic weight of the anhydrous acid = 625.0; its saturating capacity = 16.000. Its rational formula =  $C_2H_2.C_2O_3.HO$ .

*Combinations*.—With alkalis this acid forms neutral and acid salts, which are soluble and crystallizable; with earths, it forms only neutral salts; and with the oxides of the heavy metals it forms neutral and basic salts, some of which are soluble and others insoluble.

*Succinamide*,  $H_2N.C_4H_2O_2$ , is formed by the action of ammonia on succinate of oxide of ethyl; it occurs in the form of granular crystals, insoluble in cold water; like all the amides, it is decomposed by alkalis or stronger acids into the corresponding acid and ammonia.

*Bisuccinamide*, or *Succinimide*,  $C_8H_6NO_4$ , is formed on submitting succinamide to dry distillation, or on bringing dry ammoniacal gas in contact with anhydrous succinic acid; it is a white, crystallizable, fusible, soluble body, which, on being boiled with a solution of potash, takes up 2 atoms of water, and becomes decomposed into ammonia and succinic acid ( $HN.C_4H_2O_4 + 2HO = H_3N + C_8H_4O_6$ ).

*Preparation*.—This acid was, as its name implies, originally obtained from the dry distillation of amber. It was discovered in the sixteenth century. It has since been found to exist, preformed, in certain kinds of turpentine and in certain plants. It, however, occurs much more frequently as a product of the decomposition of fats, as wax, stearic acid, spermaceti, margaric acid, &c., and in various kinds of fermentation; thus, for instance, malate of lime, in contact with nitrogenous bodies, becomes gradually converted into succinate of lime ( $CaO.C_4H_2O_4 - O = CaO.C_4H_2O_3$ ).

<sup>1</sup> [Succinic acid has recently been detected by Heintz, in a cyst containing *Echinococci* in the liver. See *Jenaische Ann. f. Physiol. u. Med.* Bd. 2, S. 180, and *Poggenдорff's Ann.* Bd. 80, S. 118; or *Chemical Gazette*, vol. 7, p. 477, and vol. 8, p. 425.—G. E. D.]

According to C. Schmidt,<sup>1</sup> succinic acid is found in greater or lesser quantity in all fermented fluids, and it is possible that it is formed from glucose, together with mannite ( $C_{12}H_{12}O_{12} = C_8H_9O_8 + C_4H_2O_3 \cdot HO$ , Liebig).<sup>2</sup> This acid is usually obtained by the distillation of amber, to which a little sulphuric acid has been added; the sublimate is then purified by boiling with nitric acid.

*Tests.*—As this acid exhibits no very characteristic reactions towards other bodies, we can only determine its presence by separating it in a state of purity and then analyzing it.

#### SEBACIC ACID.— $C_{10}H_{18}O_4 \cdot HO$ .

*Properties.*—This acid (known also as *pyrolic acid*) is, in its external appearance, very similar to benzoic acid, forming white, nacreous, acicular crystals, grouped together in loose heaps: the microscope, however, readily reveals the difference in the external characters of these two acids. It forms either whorled clusters similar to margaric acid, or large plates extending from a centre, and intersecting one another at various angles, which run into sharp points, without forming an angle capable of measurement: in their mode of grouping, these crystals most closely resemble well-formed crystals of margaric acid; the individual crystalline leaflets are, however, far greater. This acid fuses at  $127^\circ$ , without losing its basic water, into a colorless oil, which, on cooling, solidifies into a crystalline mass; at a higher temperature it sublimes undecomposed; it is only slightly soluble in cold water, but in hot water, as well as in alcohol and ether, it dissolves readily; it has a pungent rather than an acid taste, and reddens litmus. By prolonged boiling with nitric acid of 1.4 specific gravity, it is gradually (in six or eight days) converted into pyrotartaric acid (C. Schlieper).<sup>3</sup>

*Composition.*—According to the above formula it consists of:

Carbon,	.	.	.	10 atoms,	.	.	.	59.406
Hydrogen,	.	.	.	8 "	.	.	.	7.921
Oxygen,	.	.	.	8 "	.	.	.	23.762
Water,	.	.	.	1 "	.	.	.	8.911
100.000								

The atomic weight of the hypothetical anhydrous acid = 1150; its saturating capacity = 8.696; its rational formula is  $C_8H_8 \cdot C_2O_3 \cdot HO$ .

*Combinations.*—Its salts are very similar to those of benzoic acid; the alkaline salts are very soluble, the earthy salts are difficult of solution, while those of the oxides of the heavy metals are insoluble.

*Pyrotartaric acid*,  $C_5H_3O_3 \cdot HO$ , is formed when nitric acid acts on sebacic acid, each atom of the latter assimilating 5 atoms of oxygen, thus  $C_{10}H_8O_8 + 5O = 2(C_5H_3O_3 \cdot HO)$ ; it is crystallizable, white, resists the action of the air, fuses at a little above  $100^\circ$ , and sublimes at a higher tempera-

<sup>1</sup> Handwörterbuch der Chemie, von Liebig, Wöhler, u. Poggendorff. Bd. 3, S. 224.

<sup>2</sup> Ibid. Bd. 3, S. 124.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 70, S. 121–129.

ture, developing at the same time a white suffocating vapor; it has a strongly acid taste, dissolves readily in water, alcohol, and ether, and in sulphuric acid without blackening, and expels carbonic acid from its salt; most of its salts are soluble in water and in spirit of wine; with neutral acetate of lead it yields no precipitate, but with the basic acetate, and with nitrate of silver, we have a white, gelatinous deposit which, on drying, becomes brownish-white, and translucent. This acid is isomeric, or probably identical, with the *lipic acid* which has been examined by Laurent and Bromeis, and is mentioned in page 76; hence it belongs to the same group of acids as sebacic acid.

*Preparation.*—This acid is formed during the dry distillation of oleic acid. As it is produced from no other kind of fat, we may determine the presence and amount of olein in a fat, from the presence and amount of the sebacic acid. In order to prepare it, the distillate must be boiled with water as long as crystals continue to be deposited from it on cooling. By a repetition of the crystallization, the acid may be obtained in a state of purity.

*Tests.*—In this distillation scarcely any other acid can occur which could be confounded with sebacic acid. It can be distinguished from benzoic acid, to which, as we have observed, it is very similar, by the circumstances that there is a precipitate on the addition of nitrate of silver or of one of the salts of the suboxide of mercury to its hot solution (which is not the case with benzoic acid); that the sublimed acid crystallizes far less readily; that a microscopic examination of the crystals obtained from the aqueous solution, reveals a difference of form; and finally that by the action of nitric acid it is converted into lipic acid.

## THE BENZOIC ACID GROUP.



This is also a group of acids which has little relation to animal chemistry, and to which we should make no reference in this place, if it were not that their representative, *benzoic acid*, sometimes occurs in animal fluids, and that its conversion in the animal body has already thrown much light on the metamorphosis of the tissues.

In accordance with the above general formula we have the following acids belonging to this group:

Benzoic acid,	.	.	.	.	.	.	.	.	$=C_{14}H_5O_3.HO,$
Myroxyllic acid,	:	:	:	:	:	:	:	:	$=C_{15}H_6O_3.HO,$
Toluylic acid,	.	.	.	.	.	.	.	.	$=C_{16}H_7O_3.HO,$
Cumic acid,	.	.	.	.	.	.	.	.	$=C_{20}H_{11}O_3.HO,$
and Copaivic acid,	.	.	.	.	.	.	.	.	$=C_{40}H_{31}O_3.HO.$

but there are certain other acids, as, for instance, *cinnamic acid*,  $C_{18}H_7O_3.HO$ , which, partly from their physical properties, and partly on account of the analogy of the products of decomposition, must be regarded as homologous to these acids, although the ratio of the carbon



to the hydrogen is not in accordance with the above formula. Moreover, we are acquainted with certain higher stages of oxidation of the same radical, to which stages we assign specific names, and which are impressed with the general character of this group. They contain 5 atoms of oxygen, and are

Salicylic acid, . . . . .	$C_{14}H_5 O_5.HO$ , corresponding to	Benzoic acid,
Anisic acid, . . . . .	$C_{16}H_7 O_5.HO$ , “	Toluylic acid,
Cumaric acid, . . . . .	$C_{18}H_9 O_5.HO$ , “	Cinnamic acid,
and Copalic acid, . . . . .	$C_{40}H_{31}O_5.HO$ , “	Copaivic acid.

All these acids have the following properties in common: they are solid, crystallize readily in needles or scales, are devoid of odor when pure, are fusible, sublime without decomposition, and are slightly soluble in cold water; they dissolve freely in hot water, and crystallize as the solution cools; they are readily soluble in alcohol and ether, and they redden litmus. Their salts present the same analogies.

Physiology itself shows us that cinnamic acid, although not constituted in accordance with the above formula, should be included in this group, for Marchand<sup>1</sup> has experimentally proved that cinnamic acid, like benzoic acid, is converted in the animal body into hippuric acid.

Hypotheses of the most varied kinds, chiefly grounded on the products of decomposition, have been set up regarding the rational constitution of these acids. These hypotheses are, however, for the most part limited to the constitution of benzoic acid, and as but few of them are applicable to the other members of this group, we may regard this as an evidence of their untenability. This is partially the case with the hypothesis of Fehling, who, previously to Kolbe, regarded benzoic acid as a conjugated oxalic acid, whose adjunct was phenyl,  $C_{12}H_5$ . Hitherto, however, the evidence in favor of any one of these hypotheses has not been sufficiently preponderating to warrant its unconditional acceptance.

All these bodies present an analogy in their relations of combination and decomposition. Thus each of these acids presents a series of lower stages of oxidation not dissimilar to the aldehydes of the first group, and containing 1 atom of hydrogen more and 1 atom of oxygen less than the corresponding acid in the anhydrous state.\* These lower oxides are sometimes acid, sometimes basic, sometimes indifferent volatile oils, some of which occur preformed in the vegetable kingdom.

Volatile oil of bitter almonds, . . . . .	$C_{14}H_6 O_2$ , corresponds with	benzoic acid, . . . . .	$C_{14}H_5 O_3$ .
Salicylous acid, . . . . .	$C_{14}H_6 O_4$ , “	salicylic acid, . . . . .	$C_{14}H_5 O_5$ .
Hydride of cinnamyl, . . . . .	$C_{18}H_8 O_2$ , “	cinnamic acid, . . . . .	$C_{18}H_7 O_3$ .
Cumin, . . . . .	$C_{18}H_8 O_4$ , “	cumaric acid, . . . . .	$C_{18}H_7 O_5$ .
Cumin, . . . . .	$C_{20}H_{12}O_2$ , “	cumic acid, . . . . .	$C_{20}H_{11}O_3$ .

In all these combinations 1 equivalent of hydrogen may be replaced by 1 equivalent of chlorine, bromine, iodine, or sulphur.

From the chlorine-combinations of this class, we can obtain the corresponding amides by the action of ammonia; thus, for instance, in the case of benzamide, the action is shown by the equation,  $C_{14}H_5O_2Cl + H_3N = HCl + H_2N.C_{14}H_5O_2$ .

<sup>1</sup> Journ. f. pr. Ch. Bd. 18, S. 35.

On submitting to dry distillation the ammonia-salts of the acids containing 3 atoms of oxygen we obtain the corresponding nitriles, which, like the nitriles of the first group of acids, are volatile, inflammable fluids. They are likewise decomposed both by strong acids and alkalis into ammonia and the corresponding acid, and when heated with potassium they yield cyanide of potassium and carbo-hydrogens.

The hydrates of the acids containing 3 atoms of oxygen, when heated with caustic alkalis, lime, or baryta, yield to them 2 atoms of carbonic acid, and become converted into non-oxygenous oils :

Hydrated benzoic acid, . . .  $C_{14}H_6O_4 - 2CO_2 = C_{12}H_6$  = Benzole or Benzin,  
 Hydrated cumic acid, . . .  $C_{20}H_{12}O_4 - 2CO_2 = C_{18}H_{12}$  = Cumole or Cumin,  
 Hydrated toluyllic acid, . . .  $C_{16}H_8O_4 - 2CO_2 = C_{14}H_8$  = Toluole or Toluin.

In these carbo-hydrogens we may again replace 1 equivalent of hydrogen by 1 equivalent of chlorine, bromine, iodine, or hyponitric acid ( $HO_4$ ); and in this way there are formed, for instance, chlorobenzide,  $C_{12}H_5Cl$ , bromocumide,  $C_{18}H_{11}Br$ , iodotoluide,  $C_{14}H_7I$ , and nitrobenzide, nitrocumide, and nitrotoluide,  $C_{12}H_5NO_4$ ,  $C_{18}H_{11}NO_4$  and  $C_{14}H_7NO_4$ .

These last-named nitrogenous compounds form yellow, oleaginous bodies, from which, by the action of sulphuretted hydrogen, we obtain the organic, non-oxygenous, volatile bases, benzidine,  $C_{12}H_7N$ , cumidine,  $C_{18}H_{13}N$ , and toluidine,  $C_{14}H_9N$  (according to the equation  $C_{14}H_7NO_4 + 6HS = 4HO + 6S + C_{14}H_9N$ ).

## BENZOIC ACID.— $C_{14}H_5O_3.HO$ .

### Chemical Relations.

*Properties.*—In its sublimed state this acid occurs in colorless, delicate needles; in the moist way it crystallizes in scales, or small prisms, or six-sided needles (the primary form of the right rhombic prism); it

Fig. 2.



Benzoic Acid.

fuses at a temperature exceeding  $120^\circ$ , boils at  $239^\circ$ , and then becomes converted into a thick, irritating vapor; it is not decomposed either by

nitric or by sulphuric acid; in other respects it has the general properties of the acids of this group.

*Composition.*—In accordance with the above formula, it consists of:

Carbon,	.	.	.	.	.	14 atoms,	.	.	.	68·858
Hydrogen,	.	.	.	.	.	5 "	.	.	.	4·098
Oxygen,	.	.	.	.	.	3 "	.	.	.	19·672
Water,	.	.	.	.	.	1 "	.	.	.	7·377
										100·000

The atomic weight of the hypothetical anhydrous acid=1412·5, and its saturating capacity=7·079.

*Combinations.*—Most of the benzoates are soluble in water; the alkaline and magnesian salts are very soluble, but do not readily crystallize; the combinations of benzoic acid with the oxides of the heavy metals are for the most part difficult of solution, but are taken up more freely by hot than by cold water.

*Products of its metamorphosis.*—*Oil of bitter almonds* is usually regarded as a combination of a hypothetical oxygenous radical (benzoyl) with hydrogen; it is thus a hydride of benzoyl,  $C_{14}H_5O_2.H$ ; it is a thin, colorless liquid whose specific gravity is 1·043 and whose boiling-point is  $180^\circ$ ; when exposed to the air it oxidizes and becomes converted into hydrated benzoic acid. It not only occurs in oil of bitter almonds, but is often found as a product of decomposition when albuminous or gelatinous substances are treated with strong oxidizing agents (Guckelberger).<sup>1</sup> The one equivalent of hydrogen of this body may not only be replaced by chlorine, bromine, or iodine, but also by sulphur or cyanogen.

*Benzamide*,  $H_2N.C_{14}H_5O_2$ , whose preparation is noticed in the introductory remarks on this group, is a beautifully crystallizable body which is soluble in water, alcohol, and ether, and possesses all the known properties of the amides.

*Benzonitrile*,  $C_{14}H_5N$ , whose formation has also been alluded to, is a colorless oil, which boils at  $191^\circ$ , dissolves in 100 parts of boiling water, and in alcohol and ether in every proportion; as, when treated with potassium, it yields cyanide of potassium, many regard it as cyanide of phenyl,  $C_{12}H_5.C_2N$ .

If azobenzide,  $C_{12}H_4N_2$ , be dissolved in alcohol, the solution saturated with ammonia, and sulphuretted hydrogen passed through it, we obtain the organic base, *benzidine*,  $C_{12}H_6N_2$ .

*Benzoin*,  $C_{14}H_6O_2$ , (isomeric with oil of bitter almonds) is formed by the contact-action of the caustic alkalies on oil of bitter almonds containing hydrocyanic acid; it occurs in prisms, which are devoid of color, taste, and smell, and which may be sublimed without undergoing decomposition; it dissolves in concentrated sulphuric acid, and in an alcoholic solution of caustic potash, communicating in each case a blue tint to the mixture; on passing its vapor through a red-hot tube it is again converted into oil of bitter almonds. By the action of chlorine it loses 1 equiv. of hydrogen, and is converted into *benzile*,  $C_{14}H_5O_2$ , which is isomeric with the hypothetical radical, benzoyl, crystallizes in sulphur-yellow six-sided prisms, and is fusible and capable of sublimation.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 64, S. 46 ff.

*Benzine* or *benzol*,  $C_{12}H_6$ , is obtained, as has been already mentioned, on treating benzoic acid with an excess of hydrated lime; it is a colorless inflammable fluid with an ethereal odor, is solid at  $0^\circ$ , boils at  $86^\circ$ , is insoluble in water, but dissolves in alcohol and ether. Amongst the many other substances which have been obtained from benzine, we may mention *nitrobenzide*,  $C_{12}H_5NO$ , a yellow fluid with a sweetish taste and a cinnamon-like odor, which is not decomposed by the alkalis. If an alcoholic solution of this nitrobenzide be treated with hydrated potash and then distilled, there is produced a non-oxygenous, nitrogenous body, *azobenzide*,  $C_{12}H_4N$ , forming large, red, fusible, and volatile crystals, which neither corresponds with the nitriles nor possesses basic properties like the organic, non-oxygenous bases.

*Preparation.*—Benzoic acid is found in many of the resins or balsams, but occurs in the largest quantity in the resin known as gum-benzoin, from which it is ordinarily prepared either by sublimation, or, in the moist way, by dissolving the resin in spirit of wine, adding an aqueous solution of carbonate of soda, and then precipitating the benzoic acid by the addition of hydrochloric acid to the filtered and concentrated fluid.

*Tests.*—Benzoic acid is less to be distinguished from other substances by its volatility, than by its property of separating in crystalline scales from very concentrated aqueous solutions on the addition of an acid. But in carrying on investigations in relation to benzoic acid we must be especially careful respecting the evaporation of the fluid, since it volatilizes very readily with the steam; we may easily perceive delicate crystals on the paper covering of the evaporating basin, when acid fluids of this nature have been evaporated without due care; it is therefore better not to add an acid to the fluid till after evaporation, or if it be already acid, to render it alkaline previously to evaporating it. I have found the following method applicable to the discovery of small quantities of benzoic acid in the animal fluids: the alcoholic extract of the fluid in question (for the alkaline benzoates and benzoate of lime are soluble in alcohol) must be mixed with a little acetic, hydrochloric, or lactic acid; if distinct crystals of benzoic acid do not now separate, the mass must be extracted with ether, and the ethereal solution submitted to spontaneous evaporation; from this ethereal extract, which is usually of an oily fluid character, the benzoic acid separates in a crystalline form on the addition of water. When too much fat is present, we must treat the separated mass with dilute spirit, which dissolves the benzoic acid without acting on the fat; on the evaporation of this spirituous solution, we obtain the benzoic acid in a tolerably pure crystalline state, mixed with other free but fluid acids. Under the microscope it appears in rectangular tablets, which, for the most part, are arrayed in rows, being linked together by their opposite angles. Its slight solubility in water, the facility with which it sublims (as may be seen with a minute quantity between two pieces of flat glass or shallow watch-glasses), together with its crystalline form, afford strong presumption of its presence. Since the remaining acids of this group, which in other respects are very similar to benzoic acid, are not found in the animal body, they cannot give rise to any confusion or mistake in testing for this acid. We

have already explained in p. 79, how it may be distinguished from succinic acid, and from sebacic acid, which, however, can scarcely be regarded as existing preformed in the animal body. The mode of distinguishing it from hippuric acid, which closely resembles it in physical properties, will be given in a future page. If we can obtain a sufficient quantity, an elementary analysis and a determination of the atomic weight are by no means superfluous.

### *Physiological Relations.*

*Occurrence.*—In a physiological point of view, benzoic acid deserves a full consideration, although numerous experiments render it probable that it does not exist preformed in any animal fluid. No one has suspected its presence in any animal fluid but the urine; and in this, both in the case of herbivora and carnivora, it occurs very often in the place of hippuric acid. Liebig,<sup>1</sup> in his classical essay on Fermentation, Putrefaction, and Decay, attributed the occasional occurrence of benzoic acid, in place of hippuric acid, in the urine of horses, solely to a process of fermentation which the latter acid underwent when the urine began to decompose; benzoic acid being formed from it, together with other products. Subsequently,<sup>2</sup> however, he changed his opinion, believing that he had ascertained that horses, when very hardly worked, and living on insufficient fodder, discharged urine containing benzoic acid, while, under the opposite conditions, the urine contained hippuric acid. In order to ascertain which, or whether either of these views were correct, I<sup>3</sup> analyzed the urine of a large number of horses, both well-fed and half-starved, and healthy and diseased; but invariably found hippuric acid and no benzoic acid, unless when the urine had been a good deal exposed to the air at an ordinary temperature. But, on the other hand, when it had stood for some time in the stable, and began to be ammoniacal, it never contained hippuric acid, but only benzoic acid. Hence, too, it is that we so often meet with only benzoic acid in human urine, which, as it contains a far smaller proportion of hippuric acid, must be employed in larger quantities; and if some portions of it have been long exposed to the air, which can hardly be avoided, they produce such a change that only benzoic acid is found in the whole urine. Hence it appears to be the fact, as Liebig assumed, that a ferment is formed in the urine through which the nitrogenous hippuric acid is converted into benzoic acid; for if we mix a specimen of urine containing benzoic acid, whether from man or from the horse, with another specimen containing hippuric acid, on separating the acids from the mixture we almost constantly obtain benzoic acid alone, the ferment of the urine containing benzoic acid probably acting on the hippuric acid of the fresh urine even during the evaporation of the mixture. Moreover, the conversion of benzoic acid conveyed into the organism, into hippuric acid, which was invariably observed by Wöhler and Keller,<sup>4</sup> Ure,<sup>5</sup> and subsequent experi-

<sup>1</sup> Ann d. Ch. u. Pharm. Bd. 30, S. 261 ff.

<sup>2</sup> Ibid. Bd. 41, S. 272.

<sup>3</sup> Handwörterbuch d. Physiol. Bd. 2, S. 14.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 43, S. 108.

<sup>5</sup> Medico-Chirurgical Transactions. Vol. 24, p. 30.

menters, is in accordance with the idea that the former, when it occurs in the urine, is only a product of decomposition of the latter.

*Action.*—We shall return to the behavior of benzoic acid in the living animal body when we treat of hippuric acid. We will here only remark that the ingestion of this acid causes an extremely disagreeable irritation in the throat,\* and subsequently a very profuse diaphoresis; and, finally, that it is one of the very few acids which produce a marked augmentation of the acidity of the urine.

### THE LACTIC ACID GROUP.



We make a special group of these acids, although their sole representative is lactic acid. Although this acid deserves a special chapter in every work on physiological chemistry, we see good reason for classing it in a special group of acid bodies. We have already remarked (see p. 62) that in its composition lactic acid presents a close analogy to metacetonc acid; it is more than probable that many other acids exist which stand in the same relation to the individual members of the first-described group of acids, as lactic acid stands to metacetonc acid; and, in point of fact, Cahours,<sup>1</sup> and subsequently Strecker,<sup>2</sup> arrived at the discovery of some such acids by a perfectly different train of ideas from that which we have pursued. The latter, in employing Piria's method of decomposing the amide compounds (given in p. 45), with the view of ascertaining whether certain nitrogenous animal substances were amides, found two such acids constituted according to the above general formula. In treating glycine with nitrous acid, he discovered an acid= $C_4H_3O_5.HO$ , corresponding to acetic acid, and on treating leucine in a similar manner, he obtained an acid= $C_{12}H_{11}O_5.HO$ , analogous to caproic acid.

Acetic acid, . . .	$C_4H_3O_5.HO$ ,	corresponds to	glycic acid, $C_4H_3O_5.HO$ .
Metacetonc acid, .	$C_6H_5O_5.HO$ ,	"	lactic acid, $C_6H_5O_5.HO$ .
Caproic acid, . .	$C_{12}H_{11}O_5.HO$ ,	"	leucic acid, $C_{12}H_{11}O_5.HO$ .

In the decomposition of hippuric acid, according to the same method, Strecker obtained a new acid, whose composition is not in accordance with the above formula, but is very similar to that of lactic acid: it is represented by the formula  $C_{18}H_7O_7.HO$ ; hence it is analogous in its constitution to the neutral carbo-hydrates of the vegetable kingdom (starch, sugar, woody fibre); that is to say, in addition to carbon it contains hydrogen and oxygen in the exact proportions to form water. Here, too, we should place the cholesteric acid= $C_8H_4O_4.HO$ , discovered by Redtenbacher, which also presents much similarity in its characters with the above-named acids of the carbo-hydrates.

There is little to be said regarding the general properties of the acids of this group, as in truth, lactic acid is the only one of them with whose

<sup>1</sup> Compt. rend. T. 27, p. 267.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 52-55.

characteristics we are accurately acquainted. It appears, however, from Strecker's communications, that all these acids, when deprived as much as possible of water, occur as oily, non-crystallizable fluids, redden litmus strongly, undergo decomposition when heated, and form soluble and in part crystallizable compounds with bases.

### LACTIC ACID.— $C_6H_5O_6.HO$ .

#### *Chemical Relations.*

*Properties.*—In its most concentrated state lactic acid is a colorless, inodorous, thick, syrupy fluid, which cannot be solidified by the most intense cold; its specific gravity = 1.215; it dissolves readily in water, alcohol, and ether, attracts water from the atmosphere, has a strongly acid taste and reaction, decomposes when heated, and displaces not only volatile acids but even many of the stronger mineral acids from their salts. Heated with concentrated sulphuric acid, it yields almost pure carbonic oxide gas, and is converted into a substance resembling humin; it gives, however, no trace of formic acid.

*Composition.*—According to the above formula it consists of:

Carbon,	.	.	.	.	.	.	6 atoms,	.	.	.	40.000
Hydrogen,	.	.	.	.	.	.	5 "	.	.	.	5.555
Oxygen,	.	.	.	.	.	.	5 "	.	.	.	44.445
Water,	.	.	.	.	.	.	1 "	.	.	.	10.000
											100.000

The atomic weight of the hypothetical anhydrous acid = 1012.5, and its saturating capacity = 9.876.

*Combinations.*—With bases lactic acid generally forms neutral salts, all of which are soluble in water, and many in alcohol, but none in ether. Most of the lactates may be heated to 150° or 170°, and some even to 210°, without undergoing decomposition. The alkaline lactates are not crystallizable, and by the greatest concentration can only be reduced to syrupy fluids; and the same is the case with the lactates of baryta, alumina, sesquioxide of iron, and bin oxide of tin; but all other lactates crystallize with tolerable facility, and are capable of resisting the action of the atmosphere. The following peculiar relation has recently been observed in the crystallizable lactates: the lactic acid obtained from animal fluids, and that produced by the fermentation of sugar, form, with the same base, salts which present certain differences in the amount of their water of crystallization, in their degree of solubility, and in their decomposition by heat (Liebig,<sup>1</sup> Engelhardt and Maddrell,<sup>2</sup> Engelhardt.<sup>3</sup>) This is, however, a subject requiring further investigation; at least Liebig thinks that he has obtained from the acid of *Sauer-kraut* a zinc-salt which corresponds with that yielded by the muscular juice; and in my own researches, whenever I have analyzed the lactic acid of the gastric juice in combination with magnesia or zinc, I have always found it corresponding with that obtained from sugar. Engelhardt dis-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 312.

<sup>2</sup> Ibid. Bd. 63, S. 83-120.

<sup>3</sup> Ibid. Bd. 65, S. 859-866.

tinguishes the acid obtained from muscular juice as *a* lactic acid, and that produced by the fermentation of sugar as *b* lactic acid.

*Lactate of lime*,  $\text{CaO}, a \overline{\text{La}} + 4\text{HO}$ ,  $\text{CaO}, b \overline{\text{La}} + 5\text{HO}$ , occurs in the form of white, hard bodies, which under the microscope are seen crystallizing in tufts of delicate needles, each two of which are so placed in relation to the other, that collectively they resemble overlapping tufts or pencils: their form is tolerably characteristic, and they cannot be confounded with other organic lime-salts, as for instance, the butyrate. Lactate of lime loses all its water at  $100^\circ$ , and is soluble in almost every proportion in boiling water and in alcohol; the salt of the *a* lactic acid dissolves, however, in 12.4 parts of water, and that of the *b* lactic acid in 9.5 parts; both salts may be heated to  $180^\circ$  without decomposition.

A crystallographic investigation shows that the *b* lactates of magnesia, of protoxide of manganese (which is colorless or of a pale amethystine tint), of protoxide of iron (which is of a pale yellow color), of cobalt (which is of a peach-color), of nickel, and of zinc, are isomorphous, since with three atoms of water of crystallization, they form vertical prisms with horizontal terminal surfaces, or with superimposed obtuse horizontal prisms.

*Lactate of magnesia*.—The salt of *a* lactic acid contains 4 atoms of water of crystallization, and is somewhat more soluble in spirit than that of *b* lactic acid.

*Lactate of nickel* is of an apple-green tint, and is difficult of solution in cold water and in spirit; the salt of *a* lactic acid loses all three of its atoms of water at  $100^\circ$ , while that of *b* lactic acid does not part with its third atom at a lower temperature than  $130^\circ$ .

*Lactate of zinc*.—The *a* lactate of zinc contains only 2 atoms of water of crystallization, which it very slowly loses at a temperature of  $100^\circ$ ; it begins to decompose at  $150^\circ$ , is soluble in 5.7 parts of cold and 2.88 of hot water, and in 2.23 parts of alcohol; the *b* lactate loses its water of crystallization very rapidly at  $100^\circ$ , bears exposure to a temperature of  $210^\circ$  without decomposition, and dissolves in 58 parts of cold and 6 of boiling water, but is almost insoluble in alcohol. C. Schmidt,<sup>1</sup> who is the only observer who has devoted great attention to the forms of microscopic crystals with the object of diagnosing such bodies in the animal fluids, gives a very accurate description and figure of the form of lactate of zinc; he mentions the club-like shape of the crystals during their process of formation, and their curved surfaces, as especially characteristic of this salt.

*Lactate of cadmium* crystallizes in anhydrous needles, and is almost insoluble in alcohol.

*Lactate of copper* formed with the *a* lactic acid crystallizes in hard, light blue, warty masses, dissolves in 1.95 parts of cold and 1.24 of hot water, and very readily in alcohol; at  $100^\circ$  it begins slowly to lose a portion of its water, and at  $140^\circ$  it decomposes, with a separation of suboxide of copper. Lactate of copper formed with the *b* lactic acid, with 2 atoms of water of crystallization, occurs in much larger crystals of a dark blue or green tint; it dissolves in 6 parts of cold and 2.2 of

<sup>1</sup> Entwurf e. allg. Untersuchungsmeth. der Säfte u. Excr. 1846, S. 78 ff.



boiling water, in 115 parts of cold and 26 of boiling alcohol; it parts with its water very readily and perfectly, both at  $100^{\circ}$ , and *in vacuo*, and does not become decomposed at a temperature lower than  $200^{\circ}$ , when it inflames and smoulders.

*Basic lactate of protoxide of tin*,  $2\text{SnO} \cdot \overline{\text{La}}$ , is a crystalline, anhydrous powder, which is very insoluble in water, and absolutely so in alcohol.

*Lactate of suboxide of mercury*,  $\text{Hg}_2\text{O} \cdot \overline{\text{La}} + 2\text{HO}$ , forms red crystals which are difficult of solution, and which, on boiling, become decomposed into a salt of the oxide, and into metallic mercury.

*Basic lactate of protoxide of mercury*,  $2\text{HgO} \cdot \overline{\text{La}}$ , forms anhydrous glistening prisms, difficult of solution.

*Lactate of silver*,  $\text{AgO} \cdot \overline{\text{La}} + 2\text{HO}$ , occurs in needles of a silky, glistening appearance, which blacken when exposed to light. This salt is almost insoluble in cold, but dissolves very readily in hot alcohol; it decomposes at  $100^{\circ}$ ; the aqueous solution, when boiled gradually, assumes a blue tint and deposits brown flocculi.

*Products of its metamorphosis.*—*Lactide*,  $\text{C}_6\text{H}_4\text{O}_4$ . On heating the ordinary, colorless, hydrated lactic acid to  $130^{\circ}$ , water and a little lactic acid distil over, whilst there remains a yellowish-white solid substance, which is very fusible, very bitter, almost insoluble in water, but dissolves readily in alcohol and ether, and whose composition is expressed by the formula,  $\text{C}_6\text{H}_5\text{O}_5$ . This product, when boiled with water, or for a long time exposed to the atmosphere, becomes again converted into ordinary hydrated lactic acid, and with milk of lime it yields the ordinary lactate of lime (Pelouze).<sup>1</sup> If, however, either this so-called anhydrous acid or the hydrated lactic acid be heated to  $250^{\circ}$ , the products of decomposition are carbonic acid, carbonic oxide, lactide and lactone, but no carbo-hydrogen. The lactide occurs as a sublimate which must be purified by solution in boiling alcohol. It crystallizes from this fluid in white tablets which fuse at  $107^{\circ}$  and volatilize at  $250^{\circ}$ ; the fused crystals solidify on cooling, into a crystalline mass, which is devoid of odor, has a slightly acid taste, and dissolves slowly in water; its conversion into lactic acid is more rapid than that of the so-called anhydrous lactic acid.

*Lactone*,  $\text{C}_{10}\text{H}_8\text{O}_4$  (produced according to the formula  $2\text{C}_6\text{H}_5\text{O}_5 \cdot \text{HO} - [2\text{CO}_2 + 4\text{HO}] = \text{C}_{10}\text{H}_8\text{O}_4$ ) is obtained on distilling anew the fluid products of distillation of lactic acid, washing the distillate with water, and drying the insoluble portion with chloride of calcium; the pure lactone is a colorless fluid with an aromatic odor and a burning taste, which boils at  $92^{\circ}$ , and when inflamed, burns with a blue tint.

*Lactamide*,  $\text{C}_6\text{H}_7\text{NO}_4 = \text{H}_2\text{N} \cdot \text{C}_6\text{H}_5\text{O}_4$ , is formed from lactide and dry ammoniacal gas: it crystallizes in colorless, right rectangular prisms, and is decomposed into ammonia and lactic acid. This body is moreover isomeric with the powerful base, *sarcosine*, discovered by Liebig, and with the longer-known indifferent substance, *urethran*. Strecker<sup>2</sup> has discovered a body of much interest, which is isomeric with lactamide. He has termed it *alanine*; its formula is  $\text{C}_6\text{H}_7\text{NO}_4$ ; and it is prepared in the following manner:

<sup>1</sup> Compt. rend. T. 19, 1219-1227.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 75, S. 27-46.

If a mixture of 2 parts of aldehyde-ammonia and 1 part of anhydrous prussic acid be heated with an excess of aqueous hydrochloric acid, this substance is formed; it must be precipitated from the hydrochlorate of ammonia which is intermingled with it, partly by crystallization, and partly by extraction with alcohol and ether; the hydrochloric-acid compound is decomposed by hydrated oxide of lead ( $C_4H_4O_2 + C_2NH + 2HO = C_6H_7NO_4$ ).

Alanine crystallizes in nacreous, oblique rhombic prisms, or in needles united in tufts; it dissolves in 4.6 parts of water at  $17^\circ$ , is slightly soluble in cold alcohol, but insoluble in ether; the aqueous solution has an intensely sweet taste, does not react on vegetable colors, and is precipitated by no reagent. It sublimes at a temperature exceeding  $200^\circ$ , in delicate, snowy crystals. When rapidly heated it is partly decomposed; on being inflamed it burns with a violet flame. By the action of nitrous acid, alanine is decomposed into nitrogen, water, and *lactic acid*. The salts of alanine are crystallizable, and more soluble than alanine itself in water, as well as in alcohol and ether.

From this beautiful discovery of Strecker's it seems almost certain that lactic acid is formic acid coupled with aldehyde. If alanine consists of aldehyde and prussic acid, and if it is converted by nitrous acid into lactic acid, we need only assume that (as often occurs) the atoms of prussic acid are decomposed into formic acid and ammonia, and that the latter is decomposed by nitrous acid into nitrogen and water. Since, moreover, the products of decomposition of the lactates (at all events of lactate of copper), support the assumption that aldehyde pre-exists in lactic acid, this hypothesis regarding the composition of lactic acid must be regarded as well established.

*Preparation.*—Lactic acid is very often formed during the fermentation of fluids containing sugar or starch, and it might as well be maintained that there is a specific lactic fermentation, as that there is a distant acetic or butyric fermentation. Hence lactic acid is not only found in milk which is turned sour, but also in the acid waters of starch fabrics, in *sauer-kraut*, in sour cucumbers, in fermented beet-root juice, &c. (The conditions under which this conversion takes place are explained in a future part of this work under the head of "Fermentation of Milk.")

The best method of obtaining lactic acid is by exposing sugar to this kind of fermentation, under the combined influence of milk and cheese.

Bensch<sup>1</sup> has employed the following practical method of obtaining it: 6 parts of cane-sugar,  $\frac{1}{18}$ th part of tartaric acid, 8 parts of sour milk,  $\frac{1}{2}$  part of old cheese, and 3 parts of levigated chalk, are mixed with 26 parts of water, and exposed to a temperature of  $32^\circ$ . In the course of eight or ten days a semi-solid magma of lactate of lime is formed; on boiling it with 20 parts of water and  $\frac{1}{18}$ th part of caustic lime, filtering it at a boiling temperature, and slightly evaporating it, the lactate of lime separates in a few days in granules. The salt must be drained and pressed, again dissolved in twice its weight of water, decomposed with  $\frac{7}{32}$  parts of sulphuric acid, the precipitated gypsum removed by filtra-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 174-176.

tion, and the acid fluid saturated with  $\frac{3}{10}$  of carbonate of zinc. The crystallized zinc-salt must then be decomposed by sulphuretted hydrogen, and the fluid concentrated, first by warmth, and afterwards *in vacuo*: the hydrated lactic acid is finally obtained in a state of purity by solution in ether.

Liebig<sup>1</sup> prepares lactic acid from the juice of flesh, in the following manner. Flesh from which the fat has been most carefully removed, is very finely chopped, repeatedly kneaded with water, and exposed to strong pressure; the fluid thus obtained is heated till it boils, filtered to remove the coagulated matters, decomposed with baryta-water, again filtered, and very strongly concentrated by evaporation. In the course of a few days the creatin crystallizes; the milky liquid poured away from these crystals is rather more strongly concentrated; and then gradually treated with small portions of alcohol, which causes the crystallization of the inosinates of baryta and potash. The mother-liquid, after the separation of the inosinates, is evaporated, and the residue extracted with alcohol; after this alcoholic extract has stood for a considerable time crystals are formed from it, while nearly pure lactate of potash remains in the mother-liquid. To this we must add sulphuric acid or a solution of oxalic acid (containing one-third of the acid), and then precipitate the sulphate or oxalate of potash by means of alcohol. The fluid filtered from the potash-salt is treated with ether, as long as any precipitation continues; the solution is then evaporated to a syrup, and treated with half its volume of spirit and five times its volume of ether, which takes up nearly pure lactic acid.

From this we may prepare lactate of lime, whose spirituous solution must be purified by animal charcoal, and evaporated, so that the salt may crystallize; the lactic acid is then readily separated from the lime-salt by sulphuric or oxalic acid with the aid of alcohol and ether.

*Tests.*—To determine the presence of lactic acid is one of the most difficult tasks in analytical animal chemistry, as is indeed evinced by the prolonged contest that existed regarding the presence or absence of this acid in the animal organism. In order to determine its presence with certainty, it must in the first place be separated from all other organic substances, but in this lies one of the great difficulties; for there is scarcely any other acid to which foreign bodies adhere so tenaciously. Liebig's method (which we have given) of preparing lactic acid from muscular juice is one of the best means of separating this acid from animal fluids. If we are sufficiently acquainted with the properties of lactic acid and its salts, we may modify this method in many respects, which is indeed the more necessary, since, in investigations relating to animal chemistry, we rarely have so large a quantity of material to work upon as is required in accurately following the steps laid down by Liebig. From most of the other animal fluids we can rarely obtain a sufficient quantity of lactic acid to serve for an elementary analysis. Indeed it often happens that we cannot even obtain enough of a *pure* lactate to enable us to determine the atomic weight. Hence, it is very often necessary to found our decision regarding the presence of lactic acid almost entirely on the crystalline form of its salts. Although many

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 312.

of the other properties of the lactates may contribute to establish the proof of the presence of this acid, yet a crystallometric investigation, made with the aid of the microscope, can alone be regarded as approximating in certainty to an elementary analysis.

In consequence of the extremely minute quantity of lactic acid to be obtained from the animal fluids, I am in the habit of adopting the following method, which may be readily modified in particular cases, with the view of studying the forms of the different salts under the microscope. The impure lactic acid prepared from the alcoholic extract by sulphuric or oxalic acid is treated with baryta-water, and the excess of the baryta removed by carbonic acid; the solution of lactate of baryta is evaporated to the consistence of a syrup, treated with alcohol, filtered, again evaporated, and then allowed to stand for some time, in order that the other baryta-salts (for instance, the butyrate and inosinate) may crystallize; the syrup is then allowed to trickle away, or if it be not withdrawn, is dissolved in water and decomposed with a solution of gypsum; the fluid from which the sulphate of baryta has been removed by filtration is strongly concentrated, and on examining it under the microscope we can readily perceive the double brushes of lactate of lime which we have already described, in addition to crystals of gypsum. On dissolving these crystals of lactate of lime in alcohol, and adding sulphate of copper to the alcoholic solution, the fluid, after standing for some time (in order that the excess of sulphate of copper and the gypsum that is formed may separate as completely as possible) is evaporated so as to crystallize, and the crystals of lactate of copper are then microscopically examined. If, by the above process, we do not succeed in obtaining distinct and measurable crystals, we must dissolve the residue in a little water; and (in order to decompose or separate any butyric acid that may be present) we must boil it strongly, filter it, and, after concentrating it, place on it a small zinc bar. Since, as we have already mentioned, lactate of copper is far more soluble in water than lactate of zinc, the zinc very soon becomes covered with white crystals of lactate of zinc, if the fluid be sufficiently concentrated, and these crystals, if they be allowed to remain for some time, may usually be easily measured under the microscope. Distinct crystalline forms may even be distinguished with the naked eye. If, however, in consequence of the want of a Goniometer, an accurate crystallometric investigation cannot be instituted, we must precipitate the solution of the zinc-salt with a boiling solution of protochloride of tin, and allow it to stand for some time; on then making a microscopic examination, we shall find clusters of crystals whose groups are composed of thick rhombic tables lying close upon one another. When we have in this way prepared and explored the different lactates (and after some practice, tolerably small quantities are sufficient for this purpose), we hardly require to make an elementary analysis or to determine the atomic weight, to enable us to decide regarding the presence of lactic acid. [Scherer has recently published an excellent memoir "On the recognition of small quantities of lactic acid in animal matters," in the fourth volume of the *Verhandlungen der physikalisch-medicinischen Gesellschaft zu Würzburg*, 1854. After the coagulation of the albuminous matters, either by heat,

or where this is not effectual, by alcohol, the filtered fluid is evaporated to dryness, any membranes that may be formed being removed. The residue is dissolved in water, and precipitated with baryta-water, in order to remove the phosphoric acid, sulphuric acid, and the earthy phosphates. The precipitate is removed by filtration; the excess of baryta is precipitated by sulphuric acid, and the fluid again filtered. The filtrate is treated with a little sulphuric acid, and is submitted to distillation, in order to separate the volatile acids; viz., acetic, formic, butyric, and hydrochloric acids. The residue left in the retort is very much concentrated, treated with strong alcohol, and allowed to stand for some days. The sulphates of potash and soda being insoluble in alcohol, crystallize and adhere to the walls of the vessel. The acid fluid is then decanted, and, after the addition of milk of lime, the alcohol is evaporated or distilled; we then filter while still warm, and remove the excess of insoluble hydrate of lime and the sulphate of lime that has been formed, and allow the filtered neutral solution to stand for some days. If the fluid should still exhibit an alkaline reaction from the presence of dissolved hydrated lime in it, this may be removed by a current of carbonic acid gas through it and subsequent ebullition, when the lime will be thrown down as a bicarbonate."

If the quantity of lactic acid be not too small, and if there be not too much colored extractive matter, the lactate of lime usually crystallizes in a few days in wart-like clusters. If, however, these crystals do not appear, we evaporate the whole fluid to the consistence of a syrup, mix it with strong alcohol, and let it stand in a cylindrical vessel, which must be placed in a moderately warm situation. A resinous deposit, almost insoluble in cold alcohol, and consisting of a combination of lime with extractive matter, is generally soon formed. After the alcoholic solution has now become clear, we pour it into a vessel with a cover, and gradually add small quantities of ether. The lactate of lime, if present even in mere traces, separates in the form of delicate white threads and soft crystalline masses, which, after being dried upon filtering paper and recrystallized from the smallest possible quantity of hot water, may be subjected to any further investigation that may be deemed necessary.—G. E. D.]

### *Physiological Relations.*

*Occurrence.*—The doubts regarding the nature of the free acid of the *gastric juice* have given rise to a great number of investigations on this point. Prout<sup>1</sup> and Braconnot<sup>2</sup> believed that their experiments showed that the gastric juice contained no lactic acid, but only hydrochloric acid. Subsequently, I thought that I had satisfactorily proved<sup>3</sup> the existence of lactic acid in the gastric juice of various carnivorous and herbivorous animals, by obtaining from it several of the lactates, and referred the occurrence of free hydrochloric acid simply to the decomposition of the metallic chlorides by the lactic acid during the evaporation

<sup>1</sup> Phil. Trans. for 1824, p. 45.

<sup>2</sup> Ann. de Chim. T. 59, p. 348.

<sup>3</sup> First edition of this work, 1840 Bd. 1, S. 284. Bericht über d. Fortschritte der physiol. u. path. Ch. im J. 1842. Leipzig. S. 10.

or distillation of the gastric juice. Hünefeld<sup>1</sup> supported this view. A period now arrived when Liebig totally denied that lactic acid occurred in any of the animal fluids, and, consequently, in examining the gastric juice of a criminal immediately after he had been beheaded, Enderlin<sup>2</sup> was just as unable to detect lactic acid, as he has been to find carbonate of soda in the blood-ash. Blondlot,<sup>3</sup> also, in examining pure gastric juice from dogs, found no lactic acid, and ascribed the acid reaction of the fluid to acid phosphate of lime, while Lassaigne,<sup>4</sup> in opposition to this view, attempted to prove the presence of free hydrochloric acid. Subsequently, experiments have been instituted by Bernard and Barreswil,<sup>5</sup> Pelouze,<sup>6</sup> and Thomson,<sup>7</sup> which have led all these chemists to believe that they have proved the existence of lactic acid in pure gastric juice. Very recently I<sup>8</sup> prepared the lactates from a larger amount of pure gastric juice than had hitherto been employed, and obtained them in such quantities that I was enabled to make an ultimate analysis of several of them, and to determine the atomic weight, which proved that the acid of the gastric juice is perfectly identical with lactic acid. I found that pure gastric juice, even on mere evaporation *in vacuo*, undoubtedly develops hydrochloric acid (in one case it amounted to 0.125%), but that there is then always an acid residue left, which, besides free lactic acid, contains lactate of lime and alkaline chlorides; whence we may conclude that there are in the gastric juice both free lactic acid and lactates, in addition to free hydrochloric acid.

According to my observations, chloride of calcium, but not chloride of sodium (as Bernard and Barreswil maintain), is decomposed during evaporation with free lactic acid, even *in vacuo*; hence it is not surprising that pure gastric juice should develop vapors *in vacuo*, which, when passed into a solution of nitrate of silver, should form chloride of silver. I must further remark, that the lactates obtained from the pure gastric juice, as well as from the contents of the stomach, had not the composition of the *a* lactic acid, but that of the *b* lactic acid obtained from sugar. Bernard and Barreswil allege, in opposition to Prout's opinion, that pure gastric juice is rendered decidedly turbid by a drop of a dilute solution of oxalic acid, while an equal quantity of oxalic acid in a solution of lime containing only  $\frac{1}{1000}$ th part of free hydrochloric acid, causes no precipitate. Further, starch, when boiled with hydrochloric acid, loses its property of being colored blue by iodine, while lactic acid does not induce this change. On boiling a solution of a lactate with a little hydrochloric acid and starch, the properties of the last-named body remain unaffected: starch boiled with gastric juice retains the property of being colored blue by iodine.

Although Schmidt long ago communicated to me that in his experiments he was unable to find any trace of lactic acid in the gastric juice of dogs, I have as yet been unable to determine the conditions under which it occurs in the gastric juice and those under which it is absent:

<sup>1</sup> *Chémie u. Medicin.* Bd. 2, S. 81 ff.    <sup>2</sup> *Ann. d. Ch. u. Pharm.* Bd. 46, S. 128.

<sup>3</sup> *Traité analytique de la Digestion.* Paris et Nancy. 1843. p. 244.

<sup>4</sup> *Journ. de Chim. méd.* T. 10, p. 78 et 189.

<sup>5</sup> *Journ. de Pharm. et de Chim.* Janv. 1835. p. 49.

<sup>6</sup> *Compt. rend.* T. 19, p. 1227.

<sup>7</sup> *Philos. Mag.* 3d series. Vol. 26, p. 420.

<sup>8</sup> *Berichte d. Gesellschaft d. Wiss. zu Leipzig.* Bd. 1, S. 100-105.

Circumstances having prevented me from providing myself with a dog with a gastric fistula, for the purpose of repeating the experiments, I collected the gastric juice of fourteen dogs which had been condemned to death by the police; they had been fed with horseflesh some (from 8 to 16) hours before they were destroyed, and a quarter of an hour before their death they were fed with fatty bones. The stomachs of most of the dogs contained no remains of the flesh, but merely fragments of bones. The most decided evidence of the presence of lactic acid in this gastric juice was obtained from the form and the characters of its salts, as well as from its saturating capacity. Since, moreover, the conditions, observed by Schmidt were also observed in this investigation, there could be the less doubt that in this case there was present not merely free hydrochloric acid, but also a considerable quantity of free lactic acid. On treating the gastric juice with lime-water, crystals, perfectly resembling those of lactate of lime, exhibited themselves on the evaporation, in vacuo, of the portion insoluble in spirit: even if all the chloride of calcium were not extracted by alcohol, and if the transition of the acid to the magnesia, protoxide of iron, or oxide of zinc or of copper, did not allow us to recognize the acid, we might have concluded that these crystals were "hydrated chloride of calcium and lime," but the determination of the saturating capacity from the magnesian salt removed all doubt.

In accordance with my former investigations I also found less hydrochloric acid than Schmidt, namely, only 0.118%, while Schmidt never found less than 0.171% even in gastric juice which was mixed with saliva; in addition to the hydrochloric acid there was, however, also 0.391% of free lactic acid present (11.682 grammes of gastric juice saturating 0.027 of a gramme of baryta).

There can be no doubt, when we consider Schmidt's well-known accuracy as a chemist, that in the cases which he analyzed the gastric juice contained no lactic acid, and that it was replaced by free hydrochloric acid. Amongst other points, Schmidt determined the amount of chlorine in the fresh gastric juice by strong acidulation with nitric acid and by precipitation with nitrate of silver; the precipitate was free from organic matter; after the excess of silver had been removed by hydrochloric acid from the solution (which had been freed from the chloride of silver by filtration), it was evaporated to dryness, incinerated, and the bases combined with chlorine were analyzed: the amount of these bases which was found was not sufficient to saturate all the hydrochloric acid calculated from the chloride of silver that was found. If now the free acid of a quantity of the same gastric juice were saturated with a solution of caustic potash or with lime- or baryta-water, it follows that exactly so much potash, lime, or baryta was required for saturation, as had been previously calculated for the excess of hydrochloric acid above the bases in the chloride of silver; this could not have been the case if alkaline lactates had been associated with the alkaline chlorides in the gastric juice.

Various authors have assumed that alkaline lactates are present in normal *saliva*, and have referred the acid reaction which is occasionally noticed in that fluid to the presence of free lactic acid, but in the small amount of solid residue which is left by the saliva, I have never been

able to establish with certainty the presence of lactates, even when operating on considerable quantities (obtained both from man and from the horse); I had, however, an opportunity of collecting large quantities of the saliva of a patient laboring under Diabetes mellitus, and in this case I convinced myself beyond all doubt of the presence of free lactic acid.

In all the cases of Diabetes mellitus which I have observed, the saliva has had an acid reaction: associated with this symptom and with intense thirst, we sometimes find a copious secretion of saliva, which we have thus a good opportunity of analyzing. As the saliva of such patients sometimes (but not always) contains sugar, I took care that it should flow directly from the mouth into alcohol, so as to avoid any possible formation of lactic acid from the sugar. The zinc-salt which was obtained, showed very distinctly the crystalline form of the lactate.

Notwithstanding the assumed neutralizing property of the bile, the contents of the small intestines of herbivorous, carnivorous, and omnivorous animals, always exhibit an acid reaction, which, however, diminishes towards the ileum; the acid reaction is strongest in the duodenum, especially in herbivorous animals. That the acid reaction here depends on the presence of lactic acid, may be most readily shown in the horse, in whose duodenum we find lactate of lime and free lactic acid, especially after the ingestion of amylaceous food.

Whether the acid reaction of the mucous secretion of fasting animals depends on lactic acid, cannot with certainty be decided, in consequence of the small quantities in which it can be collected.

I have repeatedly allowed the contents of the duodenum of a recently killed horse (healthy, and killed either in consequence of an accident or from its being affected with malleus) to flow directly into alcohol, and after filtering the fluid while hot, and concentrating it, I have obtained a white granular sediment, which, under the microscope, exhibited the well-known double-brush form of lactate of lime: a quantity collected for analysis contained 28.97% of water, and in the anhydrous state, 25.831% of lime, 32.982% of carbon, and 4.513% of hydrogen; this salt was therefore *b* lactate of lime. The lactic acid was separated in the ordinary manner from the alcoholic solution, and the magnesian and zinc salts were crystallometrically examined and quantitatively analyzed, so that there can be no doubt regarding the existence of lactic acid in this fluid.

Tiedemann and Gmelin,<sup>1</sup> and Valentin,<sup>2</sup> attribute the acid reaction of the mucus of the small intestines to lactic acid, because this mucus, on incineration, yields an ash abounding in carbonates, which, at all events, could not be the case to such a degree, if the free acid of this mucus were a mineral acid.

Moreover, the contents of the large intestine have often an acid reaction, and indeed constantly after the use of vegetable food: in two cases in which I was able to collect large quantities of these contents from a preternatural anus in the ascending colon, I obtained quite sufficient lactic acid to test crystallometrically the zinc and magnesian salts.

<sup>1</sup> Verdauung. Bd. 1, S. 349.    <sup>2</sup> Lehrb. d. Physiol. d. Menschen. Bd. 1, S. 343.



The fluid secreted by the large intestine (and indeed by the lower portion of the ileum) has always an alkaline reaction; hence the outer parts of the contents of the large intestine are for the most part neutral or alkaline; after the use of vegetable food the inner portion is, however, always acid, as was ascertained by Steinhäuser.<sup>1</sup>

Whether lactates constantly occur in the *chyle* must for the present remain undecided. In the chyle obtained in two cases from the thoracic duct of the horse (one horse having been fed with oats two hours before he was killed, and the other with starch-balls), lactic acid was recognized with certainty.

Here, as well as in the investigation of the alcoholic extract of lymph or blood, we must be careful in reference to the salts of the fatty acids; and, consequently, after the separation of the pure lactic acid by ether, the extract should be boiled with water to remove the non-volatile fatty acids, and the solution, when cooled, should be filtered; the lactic acid should then, in the manner we have already described, be transferred to baryta, from this to oxide of copper, and from the latter to oxide of zinc, so as to separate as much as possible the volatile fatty acids. This investigation leaves no doubt regarding the existence of lactates in the chyle of horses during the digestion of amylaceous food.

No one has yet definitely established the presence of lactic acid in the *lymph*, although its presence in the fluid is by no means improbable; since, independently of the circumstance that Marchand and Colberg,<sup>2</sup> as well as Geiger and Schlossberger,<sup>3</sup> found much carbonated alkali in the ash afforded by lymph, whose albuminous constituents were removed previously to incineration, and whose reaction was scarcely, or not at all, alkaline, we cannot readily perceive in what other way than through the lymph the large quantities of the lactic acid formed in the muscles can be carried away.

The recognition of lactates in healthy *blood* is just as difficult or impossible as that of uræa in the same fluid. It is probable that we shall never obtain a positive demonstration of the existence of alkaline lactates in healthy blood by direct experiment, but the simplest induction proves that they must be present there, even if they only remain in it for a very short period. We know from numerous experiments how rapidly effete matters, and especially salts of easy solubility, are removed from the animal organism by the kidneys; we know with what extreme rapidity iodide of potassium appears in the urine after it has been swallowed; and we know that it is only on that account that uræa has not yet been detected in healthy blood (notwithstanding the assertions of certain persons), for its sojourn in the blood is so very short that the quantity occurring in that fluid at the same time is scarcely to be recognized with our present chemical appliances (Marchand).<sup>4</sup> Hence it is not surprising that the presence of lactic acid has never yet been demonstrated, with all the necessary scientific accuracy, in normal blood, especially when we consider that it is removed from the circulating fluid

<sup>1</sup> Experimenta nonnulla de sensibilitate et functionibus intestini crassi. Diss. inaug. Lips. 1842.

<sup>2</sup> Poggend. Ann. Bd. 43, S. 625.

<sup>3</sup> Arch. f. physiol. Med. Bd. 5, S. 394.

<sup>4</sup> Journ. f. prakt. Ch. Bd. 11, S. 149.

in more ways than one. The combustion of the alkaline lactates—that is to say, their conversion into alkaline carbonates—exceeds in rapidity and extent their passage into the urine. Until we can prove that the lactic acid, which is accumulated in large quantity in the muscular tissue, and is found in the chyle and in the lymph, undergoes decomposition on the spot, we must assume that it passes into the blood, and the more so because we well know that chemical analysis has not yet attained such a degree of accuracy as to enable us to demonstrate the presence of lactic acid in the blood with due scientific precision. In what other way than through the blood could the lactic acid of the chyle or the muscular fibre pass into the urine? Lactic acid, like urea, may collect abnormally in such quantities in the blood as to be capable of detection by chemical analysis. Scherer<sup>1</sup> has paid especial attention to the occurrence of lactic acid in morbid blood; he observed that, during an epidemic of puerperal fever; the blood had often an acid reaction, and, as this fluid frequently contained only free albumen and no albuminate of soda, it was clear that it must contain a free acid. Scherer certainly did not demonstrate the actual presence of lactic acid in the blood; but, as he actually separated lactic acid from the exudations which were simultaneously present, and recognized it by the form of its salts, we cannot reject his conclusion that the acid reaction of the blood was also due to lactic acid. I have only thrice observed an acid reaction of the blood, under conditions similar to those described by Scherer, namely, in a case of pyæmia in a man, and in the blood of two women (from six to ten weeks after delivery). In no case could I obtain sufficient material to demonstrate the lactic acid with certainty.

The following experiments,<sup>2</sup> instituted on myself, exemplify the rapidity with which the lactates in the blood are converted into carbonates. Within thirteen minutes after taking half an ounce of lactate of soda (calculated as dry), my urine had an alkaline reaction. Moreover, that the conversion of the alkaline salts of the organic acids into carbonates (as was first proved by Wöhler) does not take place in the *primæ viæ*, but in the blood itself, is proved by direct experiments which I made on dogs, by injecting various quantities of lactate of soda into the jugular vein; after five, and at latest after twelve minutes, the urine exhibited an alkaline reaction.

In opposition to the view that lactates exist in the blood, it has been urged that the ash of blood has not an alkaline reaction, and further, that it contains no alkaline carbonates. We have shown in another part of this work that this observation of Enderlin's has not been made or confirmed by any one who has preceded or succeeded him (see "Ash of the Blood,") but that, on careful incineration, carbonated alkali always occurs in the blood; and even if this were not the case, it would be no evidence against the presence of lactic acid, since, on incinerating the blood, there is a combustion of sulphur and phosphorus sufficient to saturate the alkali previously combined with lactic acid. Further, carbonic acid is expelled from the carbonate by ordinary phosphate of soda, which is thus converted into tribasic phosphate of soda.

<sup>1</sup> Untersuchungen zur Pathol. Würzburg, 1843. S. 147-194.

<sup>2</sup> Jahresber. 1843. S. 10.

In *exudations*—those, namely, after puerperal fever—Scherer<sup>1</sup> found both free and combined lactic acid, often in very considerable quantity. (In one case there was 0·105% of free lactic acid.) In the exudations in a case of empyema, he found albumen uncombined with soda, from which he concluded that the latter had been abstracted from the former in consequence of the presence of lactic acid.

Lactic acid, which was originally discovered by Scheele in milk, does not occur in the healthy *milk* of man and animals: it is only in an abnormal state, or after a strictly animal diet, that milk which reddens litmus and probably contains lactic acid, is secreted. It is only after exposure to the atmosphere that healthy milk acquires an acid reaction, which is dependent on the formation of lactic acid from the sugar of milk by fermentation.

It is now forty-two years since Berzelius<sup>2</sup> recognized the existence of free lactic acid in the *muscular fluid*; and no one who has repeated the experiments of this most faithful and accurate experimentalist, can confound this acid with any other, since its properties, and those of its salts, have been made known by more recent investigations. Berzelius did not deem it necessary at that time to confirm the proof of the presence of lactic acid in this fluid by an elementary analysis, although he might readily have made one. Liebig, so long as he relied on the investigations of his pupils, absolutely denied the existence of lactic acid in the living animal body; but on instituting and publishing his own admirable inquiry respecting the fluids of the muscular tissue of animals, he could no longer question its presence in the muscular fluid, and even admitted its existence in the gastric juice. Moreover, the free acid exists in so preponderating a quantity in the muscles, that Liebig is of opinion that it is more than sufficient to saturate the alkali of all the alkaline fluids of the animal body. Berzelius thought that he had convinced himself that the amount of free lactic acid in a muscle is proportional to the extent to which it has been previously exercised.

I have likewise detected lactic acid with certainty in the juice of the smooth muscles;<sup>3</sup> and Scherer has recently detected its presence in the juice of the spleen<sup>4</sup> and in leucæmic blood.<sup>5</sup> In our investigations on the sweat both Schottin and myself failed in detecting it either in the healthy or morbid fluid. Robin and Verdeil<sup>6</sup> have recently found lactate of lime in large quantity in the urine of the horse, and Lassaigne<sup>7</sup> believes that he has found lactate of soda in the allantoic fluid of a calf.

Berzelius separated the lactic acid from the alcoholic extracts of the animal fluids in the following manner. The alkalies having been precipitated by tartaric acid, the filtered acid solution was digested with carbonate of lead; the alcoholic solution of lactate of lead, having been separated from the other lead-salts by filtration, was then treated with sulphuretted hydrogen, which left the lactic acid in solution contami-

<sup>1</sup> Op. cit.

<sup>2</sup> Lehrs. d. Ch. Bd. 9, S. 578; Ann. d. Ch. u. Pharm. Bd. 1, S. 1; Jahresber. Bd. 27, S. 585-594.

<sup>3</sup> Walther, Diss. inaug. Med. Lips. 1851.

<sup>4</sup> Verhand. d. phys.-med. Ges. zu Würzburg. Bd. 2, S. 299.

<sup>5</sup> Ibid. Vol. 2, p. 324.

<sup>6</sup> Mém. de la Société de Biologie, t. i. p. 25.

<sup>7</sup> Ann. de Chim. et de Phys. 1850. t. 17, p. 295.

nated merely with extractive matter. After the evaporation of the alcohol the acid was filtered through animal charcoal, from which the earthy salts had been separated, and treated with hydrated oxide of tin, on which the comparatively insoluble lactate of tin was separated. This was again decomposed with sulphuretted hydrogen, and the lactic acid further examined.

Anselmino, Thenard, and Berzelius<sup>1</sup> believe that they have found lactic acid and lactate of ammonia in the *sweat*.

Berzelius<sup>2</sup> also conjectures that alkaline lactates exist in the *bile*.

In consequence of the rapidity with which the alkaline lactates undergo a transformation in the blood, it would naturally follow, that lactic acid, when it occurs in the *urine*, would exist there as an extremely variable constituent; and this assumption is confirmed by experience. Earnestly as I formerly maintained the view, that lactic acid constantly occurs in animal urine, and that the acid reaction of this fluid is solely dependent on its presence, I have since convinced myself that my earlier modes of analysis (when I rested satisfied with the mere exhibition of the zinc-salt), though most carefully conducted, were open to deceptions in reference to this acid; but to maintain that the urine of healthy men and animals never contains lactic acid, or lactates, under any physiological relations, is to err just as much in the opposite direction. A more extended investigation has led me to the following results. In all cases, where the supply of lactates to the blood is very great,—whether this depends on an excess of acid being formed in the muscles, or on the use of a diet tending to produce it, or on an imperfect process of oxidation in the blood,—lactic acid may be detected in the urine, with all the certainty which, in the present state of chemistry, can be expected in such researches. Hence we can understand why it is that, in the urine of the same individual, lactic acid may on one day be present and on another absent; why, in many persons, no lactic acid can be detected in the urine, and in others again (and especially in those who in consequence of repeated catarrhs suffer from partial relaxation of the pulmonary tissue, and yet often think themselves perfectly well) it is constantly present in the urine; why stall-fed animals, living on amylaceous fodder, excrete lactic acid by the kidneys (and in part also by the mammary glands), while under other conditions this acid cannot be discovered in their urine; and why, finally, in most febrile diseases, lactic acid may be recognized in the urine.

The details of these investigations, which will be given in another place, afford numerous confirmations of the experiments which I formerly instituted on the urine.<sup>3</sup> Berzelius,<sup>4</sup> during his later years, entertained no doubt regarding the correctness of the results which he had so long before obtained, in reference to the presence of lactic acid in the urine. Boussingault<sup>5</sup> has quite recently found lactic acid in the urine of pigs fed with potatoes, as well as in that of cows and horses. (In the urine of the horse he found 1.128% of lactate of potash, and 0.881% of lactate of soda.)

<sup>1</sup> Lehrb. d. Ch. Bd. 9, S. 393.

<sup>2</sup> Ibid. S. 203.

<sup>3</sup> Journ. f. prakt. Ch. Bd. 25, S. 1, and Bd. 27, S. 257; Handwörterb. d. Physiol. Bd. 2, S. 10.

<sup>4</sup> Jahresber. Bd. 27, S. 590.

<sup>5</sup> Ann. de Chim. et de Phys. 3 Sér. T. 15, p. 97–114.

In accordance with this view is the almost universal occurrence of lactic acid in urine containing a considerable quantity of oxalate of lime, so that, by a microscopic examination of a specimen of urine, a conclusion may often be drawn regarding the presence or absence of lactic acid. Hence in those diseases in which there is an increase in the amount of oxalate of lime, as in pulmonary emphysema, disturbances of the nervous system, rachitis, &c., lactic acid is always associated with this salt. Scherer<sup>1</sup> and Marchand<sup>2</sup> have sometimes observed a considerable augmentation of lactic acid in the urine in rachitic children, and I have also noticed it in the osteomalacia of adults.

In determining the presence of lactic acid, we must always employ fresh urine if we wish to draw any conclusion regarding the composition of the renal secretion. The admirable investigations of Scherer<sup>3</sup> regarding urinous fermentation were the first to direct attention to the circumstance, that there is a gradual augmentation of the free acid when the urine is exposed to the atmosphere. The lactic acid must then be formed from some unknown matter,—probably from what we term an extractive matter. I<sup>4</sup> had formerly observed something similar occur in diabetic urine, since, when freshly passed, I always found it neutral, although subsequently it became acid; in consequence, however, of diabetic urine containing sugar, these experiments were of less weight than those of Scherer. We may hence fairly conclude, that the urine, after its excretion from the kidneys, undergoes a similar acidification in the bladder; and, consequently, that the lactic acid which is often found in the urine discharged from that viscus, is a product of decomposition which is formed externally to the sphere of vital activity. If, however, the occurrence of crystals of free uric acid warrants us in inferring the existence of the lactic fermentation, it is only very seldom that it can occur in the bladder; for the cases are extremely rare in which urine, on its emission from that organ, contains free uric acid—the statement that has found its way into various books, to the effect that fresh urine often contains free uric acid, being a very erroneous one.

C. Schmidt<sup>5</sup> has separated lactic acid in the form of lactate of zinc from the strongly acid fluid yielded by the *long bones in a case of osteomalacia*. He measured the angles of the crystals, and submitted the salt to an elementary analysis.

*Origin.*—If we might be permitted to hazard a conjecture regarding the production of lactic acid from its occurrence in the animal body, we should ascribe to it a double origin. No one can entertain a doubt that the lactic acid, found in the contents of the intestines and in the chyle after the digestion of vegetables, owes its formation to the amylaceous or saccharine matters contained in the food, which in their passage through the *primæ viæ* become converted into that acid, in the same manner as takes place in the fermentation of milk. But the true genesis of the lactic acid, which accumulates in such large quantity in the muscles, is not so immediately obvious: we may certainly assume that the lactic

<sup>1</sup> Untersuchungen z. Pathol. S. 74, ff.

<sup>2</sup> Lehrbuch d. phys. Ch. S. 105.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 42, S. 171; and Unters. z. Pathol. S. 1–16.

<sup>4</sup> De urina diabetica. Diss. inaug. Lips. 1835.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 302–306.

acid formed in the *primæ viæ* from vegetables is especially attracted by some mechanical or chemical influence of the muscular fibre, and is accumulated there to serve certain definite purposes; but this view is in some measure opposed by the circumstances, that the muscles of carnivorous animals contain as much lactic acid as those of herbivorous animals, and that free lactic acid is always found in the urine of carnivora and of men, when living on a strictly animal diet, which would scarcely be the case if the acid conveyed to the muscles solely proceeded from the lactic acid contained in the flesh which had been taken as food. But if we regard the lactic acid of the juice of flesh merely as a product of metamorphosis which is formed while the muscular fibre is discharging its function (*i. e.* during the contraction of muscle), the only objection to the view, that this acid proceeds from the decomposition of the muscular substance itself, is, that hitherto lactic acid has not been produced either by fermentation or otherwise from any nitrogenous animal matter, either albuminous or gelatinous. We should, however, not make much progress in our physiological inquiries, if we set down as impossible all the processes which we happen not yet to have recognized external to the living body. Recent investigations respecting the various modes of decomposition and the products of albuminous bodies show, that a partial conversion of albuminous matter into lactic acid is by no means an absurd impossibility; for Guckelberger,<sup>1</sup> who found aldehyde among the products of oxidation of albuminous bodies, points out, that in these substances there must be hidden a group of atoms, from which sugar of milk or lactic acid might be produced. He, further proved, experimentally, that sugar of milk with chromic acid also yields aldehyde; and, on the other hand, Engelhardt found aldehyde of acetic acid among the products of distillation of lactate of copper. We have already directed attention to the analogy existing between lactic acid and that frequent product of the metamorphosis of animal matter, metacetic acid. Hence it would be not at all surprising if lactic acid were in some manner obtained from the gelatinous or protein compounds.

Moreover, this view is supported by the consideration, that, besides lactic acid, creatine, which is found in the muscular fluid, is often a product of decomposition of muscular substance, since otherwise it would be found in other places besides the urine. Moreover, according to Liebig's discovery, creatine is decomposed by alkalies into uræa and sarcosine, a substance isomeric with lactamide: hence there would be nothing incongruous in assuming, that in the natural metamorphosis of creatine in the animal body, where no sarcosine is found, the creatine is still decomposed into uræa; but that, in place of sarcosine, there is an abstraction of water, and that lactic acid and ammonia are formed—in which case, however, we should have to explain what becomes of the ammonia. Moreover, it cannot be supposed that lactic acid passes into the muscular substance from the blood, where it is so easily and rapidly consumed; yet such must be the case, if it comes from the acid formed in the intestinal canal from amylaceous food.

Finally, after the discovery made by Redtenbacher, that glycerine is convertible into metacetic acid, there seems to be something attractive

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 64, S. 99.

in the hypothesis that glycerine, which, in the metamorphosis of the fats, obviously undergoes an independent change, is converted into lactic acid, which, as we have already shown, is allied to metacetic acid. As we have no probable conjectures regarding the further course of the haloid base of the fats in the animal body, it is possible that these substances may contribute, through their base, to the formation of lactic acid.

We have endeavored, in the above sketch of the occurrence of lactic acid in the animal body, to restrict ourselves most rigidly to established facts, and we have rejected all those of our own experiments on which the slightest doubt appeared to rest; without referring to authorities, we have allowed the facts to speak for themselves, and have attached as little credit to the negative assertions of Liebig, as to the older experiments of Berzelius, regarding the occurrence of lactic acid in bile, sweat, &c., with that impartiality which becomes every one wishing to be an honest scientific observer. We shall now consider the advantages which may accrue to the animal organism from the occurrence of lactic acid in this or that organ, without any reference to the views and errors which we formerly maintained. Although we no longer regard lactic acid as one of the most important elements in relation to the metamorphosis of the animal tissues, it is yet of sufficient importance to attract the attention of physiologists. It is moreover obvious that questions regarding the function of a substance in the animal body, can never receive more than a hypothetical answer; for purposes may indeed be conjectured or understood, but they cannot be palpably demonstrated. If, therefore, we judge of the physiological importance of an animal substance on hypothetical grounds, we do not necessarily adopt lax and untenable illusions of the fancy, but shall confine ourselves to logical conclusions.

*Uses.*—In ascribing to lactic acid an essential influence on the *digestion* of nitrogenous food, our opinion is based, not on a mere conjecture derived from the constant occurrence of this acid in the gastric juice, but on the result of direct experiments<sup>1</sup> with artificial digestive fluids, from which it appears that lactic and hydrochloric acids cannot be replaced in the process of digestion, by any other animal or organic acids. The question how the acid acts, will be entered into in our observations on "Digestion."

It is not probable that the lactic acid and lactates found in the *contents of the stomach and intestines*, are entirely derived from the acid of the secreted gastric juice; indeed it is certain that the greater part of the lactic acid, occurring both there and in the chyle, may be traced to the conversion of the starch or sugar of the food; we should, however, on the other hand, be drawing too general a conclusion, if we assumed that all the starch and all the sugar of the food must be converted into lactic acid, in order that the functions of the organism may be duly fulfilled. In the course of our subsequent physiological considerations, we shall explain the grounds why we cannot accept this view, notwithstanding that it is apparently supported by positive observations. This much is, however, supported by facts, that a portion of these substances is actually converted into lactic acid, and passes into the blood in the form of alkaline lactates. If we adopt Liebig's ingenious division of food,

<sup>1</sup> Berichte der Gesellsch. der Wiss. zu Leipzig. 1849.

into true food for nutrition and food for the respiration, we know of no substitute which could better act in the blood as food for the respiration than the alkaline lactates, which, as we have seen, undergo rapid combustion in the blood, and are thus converted into carbonated alkali,—in a word, nothing could be a better supporter of animal heat than the alkaline lactates.

If the lactic acid in the fluid saturating the *muscles*, although undoubtedly derived from the effete muscular tissue, be not a pure product of decomposition, there is much in favor of Liebig's<sup>1</sup> hypothesis, that an electric tension influencing the function of the muscles, is established by the acid muscular juice and the alkaline contents of the capillaries.

In the *urine* and *sweat*, lactic acid occurs only as a product of excretion; for even if, in some cases, it may contribute to the solution of the earthy constituents of the urine, its occasional absence in this fluid shows that other substances effecting that object are also present.

I formerly regarded lactic acid as one of the most important agents in the solution and transportation of many of the animal substances and earthy salts of the animal organism; but a more thorough insight into the processes of animal chemistry, has led me almost entirely to renounce this view; for although I<sup>2</sup> have recently convinced myself that the solvent power which lactic acid exerts over basic phosphate of lime, far exceeds that of acetic acid, and is indeed very considerable—a fact long ago asserted by Berzelius,<sup>3</sup> and directly proved by the experiments of Gay Lussac,<sup>4</sup> but whose accuracy has been called in question by Liebig,<sup>5</sup>—yet I cannot overlook the circumstance that the albuminous bodies, which are never devoid of phosphate of lime, and often contain a large quantity of it, afford far better means of transport for the bone-earth in the animal body than lactic acid can do.

How far my former view, that lactic acid is the most important factor in the metamorphosis of the animal tissues, can still be maintained, may be seen from the preceding observations.

#### SOLID FATTY ACIDS.



From this formula it is obvious that these acids stand in a close alliance with those which we have described in the commencement of this work;—indeed, we have already associated them with the latter in a single group, to which we have applied the name of fatty acids; but we meet here with the same difficulties which present themselves in inorganic chemistry, in the definition and classification of the metals. Nature recognizes no limits corresponding with our artificial systems, but for the purposes of study a separation or arrangement is always useful, provided it be not altogether at variance with nature. These fatty acids have, however, certain essential characters, which distinctly separate them from the first-named acids. Independently of the high atomic weight of the acids we are now considering, and of the circumstance that a very differently constituted group of fluid acids is closely allied to them, the following are the properties which characterize them

<sup>1</sup> Op. cit.

<sup>2</sup> Jahresh. der ges. Med. 1848, S. 10.

<sup>3</sup> Lehrb. d. Ch Bd. 9, S. 423.

<sup>4</sup> Pogg. Ann. Bd. 31, S. 399.

<sup>5</sup> Chemie in Anwendg. f. Physiologie.



as a special group. At an ordinary temperature they are solid, white, and crystalline, devoid of smell and taste, leave on paper a fatty spot which does not disappear, are lighter than water, fuse below  $100^{\circ}$ , can only be distilled unchanged *in vacuo*, are perfectly insoluble in water, dissolve in boiling alcohol, and again separate from it in crystalline forms as the solution cools, dissolve readily in ether, decompose when heated in the air, and are inflammable; their alcoholic solution only faintly reddens litmus; with a gentle heat they expel carbonic acid from its salts; with most bases they form insoluble salts (the alkaline salts alone being soluble in water), and they have a strong tendency to form acid salts with bases.

Very few of these acids have been found in the animal body; one of them, however, *margaric acid*, is the principal constituent of all the fats yet found in the animal body. Associated with it is another fatty acid, *stearic acid*, whose composition, although not in accordance with the above formula, approximates so nearly to it that it may be regarded as produced from 2 equivalents of margaric acid, from which 1 equivalent of oxygen has been abstracted. We place before our readers the whole group of these acids with their chemical formulæ, restricting, however, our observations, to the two above-named acids.

Cocinic acid, . . . . .	$C_{22}H_{42}O_4.HO.$
Laurostearic acid, . . . . .	$C_{24}H_{48}O_4.HO.$
Myristic acid, . . . . .	$C_{28}H_{56}O_4.HO.$
Palmitonic acid, . . . . .	$C_{31}H_{58}O_4.HO.$
Palmitic acid, . . . . .	$C_{32}H_{64}O_4.HO.$
Bogic acid, . . . . .	$C_{33}H_{66}O_4.HO.$
Margaric acid, . . . . .	$C_{31}H_{62}O_4.HO.$
Cocostearic acid, . . . . .	$C_{35}H_{70}O_4.HO.$
Behenic acid, . . . . .	$C_{42}H_{84}O_4.HO.$
Cerotic acid, . . . . .	$C_{54}H_{108}O_4.HO.$
Stearic acid, . . . . .	$C_{68}H_{136}O_5.2HO=2C_{34}H_{68}O_3.HO-O.$

### MARGARIC ACID.— $C_{31}H_{62}O_4.HO.$

#### *Chemical Relations.*

*Properties.*—This acid has all the properties which we have enumerated above as pertaining to this group. It crystallizes from a hot alcoholic solution in groups of very delicate macreous needles, which under the microscope appear interlaced like tufts of grass, and arranged in ensiform plates, or grouped in star-like forms. The acid, when thoroughly dried, fuses at  $56^{\circ}$ ; even when most carefully heated *in vacuo*, it can only be partially distilled unchanged, carbonic acid and margarone ( $C_{33}H_{66}O$ ) being always formed; by prolonged contact with nitric acid, it becomes finally decomposed into succinic, suberic, and carbonic acids, and water.

*Composition.*—According to the above formula this acid contains:

Carbon, . . . . .	84 atoms,	. . . . .	75.556
Hydrogen, . . . . .	88 "	. . . . .	12.222
Oxygen, . . . . .	8 "	. . . . .	8.889
Water, . . . . .	1 "	. . . . .	8.833
<hr/>			
			100.000

The atomic weight of the hypothetical anhydrous acid = 3262.5, and its saturating capacity = 3.065.

*Combinations.*—Margaric acid forms both neutral and acid compounds with *alkalies*; the acid salts are principally formed by the addition of much water to the neutral salts; with *oxide of lead* it forms acid, neutral, and basic, salts, all of which are soluble in petroleum and oil of turpentine, and the first two in heated alcohol.

*Margaramide*,  $\text{H}_2\text{N.C}_{34}\text{H}_{33}\text{O}_2$ , is formed when *olive oil* is digested in alcohol saturated with ammonia; it crystallizes in fine, silky, glistening needles, is insoluble in water, and is more soluble in hot alcohol and other than in cold, from which it separates in glistening plates; it fuses at  $60^\circ$ , and when ignited, burns like tallow.

On treating margaric acid with peroxide of lead, Bromeis<sup>1</sup> obtained a fatty acid which separated in granules and contained 1 atom more of oxygen than margaric acid; its composition being represented by the formula  $\text{C}_{34}\text{H}_{33}\text{O}_4\cdot\text{HO}$ .

*Preparation.*—Since margaric acid, in the compound which we call margarin, occurs in almost all vegetable fats (the fatty oils) as well as in the most common animal fats, it may be prepared from any of these sources. The best method of obtaining it is to take the fat of man or of the pig, or a vegetable fat, and to saponify it with potash so as to form a clear, viscid, soapy solution; this must be treated with sulphuric acid, which causes a separation of a mixture of stearic, margaric, and oleic acid; this fatty mass must be then well washed with water, dried as thoroughly as possible, and strongly pressed between paper, which causes the removal of a great part of the oleic acid. The solid acids must now be recrystallized in alcohol. The stearic acid is the first to separate from the hot alcoholic solution, and it thus admits of separation and removal; the margaric acid always separates somewhat later; in order, however, that the stearic acid may be perfectly removed, this process must be several times repeated.

We thus obtain margaric acid with no impurity beyond a little oleic acid, which may be removed by saturating the acids with an alkali and precipitating with acetate of lead; as the oleate of lead is soluble in boiling ether, while the margarate of lead is insoluble, we have an easy means of separating the two salts. The margarate of lead must then be decomposed by an alkaline carbonate, and the resulting alkaline salt by a stronger acid. The margaric acid which is thus separated may be further purified by solution in hot alcohol.

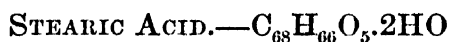
*Tests.*—From the properties, as well as from the mode of preparing this acid, we perceive that it can only be distinguished from other similar acids when it is perfectly free from any admixture with them: we may derive some information on this head from its boiling-point; but it is only by an elementary analysis that we can arrive at any certain conclusion. In the investigation of small quantities, when a separation or an analysis is out of the question, we must trust solely in a microscopical examination, which, however, in this case yields by no means such uncertain results as is generally supposed.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 42, S. 56.

*Physiological Relations.*

*Occurrence.*—It has already been remarked that margaric acid is the principal constituent of most animal fats; but this acid is here ordinarily combined with the hypothetical haloid base, *oxide of lipyl*, which, in its separation from this and similar acids, is converted into the well-known body, glycerine. Of margarin itself we shall speak in a future part of this volume, and we shall consequently defer for the present all remarks on the physiological function of margaric acid and its organic salts. But margaric acid occurs both in a free state and in combination with alkalis in most of the animal fluids, with the exception of urine; being free in acid fluids, and in a state of combination in those with an alkaline reaction; it is always accompanied by oleic acid or its salts. Its presence in the saliva, in the blood, in exudations of all kinds, in pus, and in the bile, is so easily recognized, that it is unnecessary to quote authorities regarding its existence in these fluids; moreover, in our remarks on these fluids we shall return to this subject. We will here only remark that it may also be discovered in the solid excrements after the use of vegetable food, and that it occurs in considerable quantity in dejections which have been caused by purgatives or mineral waters. As already mentioned, we must here always have recourse to the microscope, by which, independently of any chemical process, free margaric acid may often be detected in acid pathological fluids; thus, in acid pus discharged from what are termed cold abscesses, or in pus in which acid fermentation has with all due caution been established, the most beautiful crystals of margaric acid are formed; more beautiful indeed than we could artificially prepare.

We shall postpone our observations regarding the *origin* of margaric acid in the animal organism, and the *rank* and *position* it holds in the metamorphosis of the animal tissues, till we take into consideration the formation and the physiological importance of the fats in the animal body.

*Chemical Relations.*

*Properties.*—This acid crystallizes in white, glistening needles or leaflets, which, however, under the microscope, appear as very elongated, lozenge-shaped plates, with the obtuse angles rounded off, as in the microscopical whetstone-like crystals of uric acid; these crystals are, however, much longer, and have a far shorter transverse diameter than the similar crystals of uric acid. They often collect at one spot, the acute angles slightly overlapping one another, so that when seen under the microscope the crystals present the arrangement of whorl-shaped clusters. This acid begins to fuse at  $75^{\circ}$ , but again solidifies if the temperature is reduced to  $70^{\circ}$ . Submitted to a dry distillation it yields hydrated margaric acid, margarone, and an oleaginous carbo-hydrogen; by prolonged digestion with nitric or chromic acid it becomes perfectly converted into margaric acid. In the cold, stearic acid decomposes the carbonated alkalis to the amount of one-half, but with the aid of heat a perfect decomposition is effected.

*Composition.*—According to the above formula, stearic acid contains:

Carbon, . . . . .	68 atoms,	. . . . .	76.692
Hydrogen, . . . . .	66 "	. . . . .	12.406
Oxygen, . . . . .	5 "	. . . . .	7.519
Water, . . . . .	2 "	. . . . .	3.883
<hr/>			
100.000			

The atomic weight of the hypothetical dry acid = 6425: its saturating capacity (if we regard as neutral the salt containing 2 atoms of base). = 3.113.

*Combinations.*—The neutral alkaline stearates (containing 2 atoms of fixed base) dissolve unchanged in from 10 to 20 parts of water; in a very large quantity of water they become decomposed, an acid salt separating, and the fluid becoming very strongly alkaline; the alcoholic solution of the acid salt reddens litmus, but on the addition of water to this solution the reddened litmus again becomes blue. The compounds of stearic acid with all other bases are insoluble in water. For *stearate of oxide of lipyl* (or of *glycerin*) see "Stearin."

*Preparation.*—As this acid does not occur in vegetable fats, and exists only in very small quantity in most of the animal fats, except in mutton fat, it is from this last-named source that it is most advantageously prepared; we obtain it in accordance with the method indicated in our remarks on margaric acid, by boiling with alcohol of 0.83 spec. grav. the fatty acids separated by sulphuric acid from the soap; this leaves a residue of stearic acid tolerably free from margaric acid; by repeated solution in absolute alcohol it becomes purified, till we finally obtain a mass possessing the known fusing-point of this acid. The following method of preparing it may also be recommended. Dissolve saponified mutton fat in 6 parts of warm water, and then wash it well with a large quantity of cold water; a gradual separation of a glistening nacrous mass now ensues, consisting of distearate and bimargarate of potash. This must be dissolved in 20 times its bulk of hot alcohol, from which, as it cools, the stearate alone separates; on decomposing this salt with hydrochloric acid, the free acid may be obtained by remelting it in water.

*Tests.*—An elementary analysis can only be instituted as a test for the presence of stearic acid, when there is a sufficiently large quantity of fat present to admit of the above-mentioned separation of stearic and margaric acids,—a separation which, unfortunately, is only practicable when we have very large quantities to deal with. Hence this, the most certain method, is only applicable in determining the amount of stearin in an animal fat. In dealing with smaller quantities we must rest content with the microscopic investigation of the fatty acids separated from hot alcoholic solutions. In order to obtain a scale for the approximate ratios of a mixture of margaric and stearic acids, Gottlieb<sup>1</sup> has determined the fusing-points of various mixtures of these acids. His results are as follows:

	Stearic acid.		Margaric acid.	Fusing-point.
1)	. . . . . 30 parts,	to	10 parts,	. . . . . 65°·5
2)	. . . . . 25 "	"	10 "	. . . . . 65°
3)	. . . . . 20 "	"	10 "	. . . . . 64°
4)	. . . . . 15 "	"	10 "	. . . . . 61°
5)	. . . . . 10 "	"	10 "	. . . . . 58°
6)	. . . . . 10 "	"	15 "	. . . . . 57°
7)	. . . . . 10 "	"	20 "	. . . . . 56°
8)	. . . . . 10 "	"	25 "	. . . . . 56°·5
9)	. . . . . 10 "	"	30 "	. . . . . 56°·3

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 35.

Both pure margaric and pure stearic acids, after having been fused and again allowed to solidify, are perfectly crystalline; stearic acid, however, forms small confused crystals, while margaric acid forms larger acicular crystals; a mixture of the two acids is, however, in this state, far less crystalline, and presents rather a porcelain-like, opaque, and brittle appearance.

### *Physiological Relations.*

*Occurrence.*—Like margaric acid, stearic acid occurs in most animal fats; it is, however, always found in less quantity than margaric acid, and in some cases appears to be altogether absent; or, at least, our present chemical appliances fail in detecting it. In the fat of the cellular tissue it exists like margaric acid in combination with glycerine; it never occurs free unless in association with margaric acid; it is, however, of much rarer occurrence than free margaric acid, and occurs in much smaller quantity.

*Origin.*—As stearic acid is never found in vegetable fats, it must be primarily formed in the animal body, where, indeed, its formation may be readily explained. As it consists of 2 atoms of margaric acid *minus* 1 atom of oxygen, we may regard it as produced from margaric acid, to which it stands, as we have seen, in the same relation as hyposulphuric acid to sulphuric acid, for  $S_2O_5 : SO_3 = (C_{34}H_{53})_2O_5 : (C_{34}H_{53})O_3$ .

In which part of the system this conversion occurs we do not at present know: that it takes place in the blood is improbable, because we assume that the fats are directly oxidized in the blood, and are decomposed into the oxides of simpler radicals. That this conversion takes place in the *primæ viæ* is, at all events, incapable of demonstration.

We shall speak of the uses of stearic acid in the animal organism, in our remarks on the fats in general.

### OILY FATTY ACIDS.



This group of bodies contains a far smaller number of members than the preceding groups. At present the following are the only oily fatty acids with which we are acquainted:

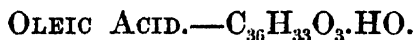
Oleic acid,	.	.	.	.	.	.	.	$C_{36}H_{53}O_3.HO.$
Erusic acid,	.	.	.	.	.	.	.	$C_{44}H_{67}O_3.HO.$
Doeglic acid,	.	.	.	.	.	.	.	$C_{38}H_{55}O_3.HO.$

*Ricinoleic acid*, containing the same group of atoms of carbon and hydrogen with 5 atoms of oxygen ( $=C_{38}H_{55}O_5.HO$ ), bears the same ratio to the last of these acids, which salicylic acid bears to benzoic acid.

Dissimilar as, on the whole, is the composition of the oily and the solid fatty acids, they are yet similar in most of their physical and even in many of their chemical properties.

Whether *campholic acid*  $C_{20}H_{17}O_3.HO$ , and the two isomeric acids, *campheric acid* and *angelic acid*  $=C_{10}H_7O_3.HO$ , belong to this group (for their composition accords with the general formula  $C_mH_{m-3}O_3.HO$ ) is as yet undecided; several of their physical properties (for instance, they

are solid, crystallizable, and volatile) do not accord with this view, but these acids may possibly bear the same relation to the oily acids, that the acids of the first group bear to the solid fatty acids, and the low atomic weight of the radical may also be the cause of this difference in their properties.



#### *Chemical Relations.*

*Properties.*—This body, known also as elaic acid, is, when perfectly pure, and at a temperature above  $+14^\circ$ , of an oily consistence, limpid, devoid of color, taste, and smell, and exerts no action on litmus; at  $+4^\circ$  it forms a white, crystalline mass, which, at the moment when it solidifies, strongly contracts and expresses the still oily portion; it is then very hard, and is unaffected by exposure to the atmosphere; on exposing an alcoholic solution to extreme cold it crystallizes in long needles. In its fluid condition, that is to say, as oil, it rapidly absorbs oxygen and becomes changed. When heated, it becomes decomposed, yielding not only carbon, carbonic acid, and carbo-hydrogens, but capric and caprylic acids, and especially sebacic acid. Finally, on treating oleic acid with hyponitric acid, the whole mass becomes solid and converted into elaidic acid. By prolonged treatment with nitric acid, oleic acid yields (according to Laurent<sup>1</sup> and Bromeis<sup>2</sup>) the acids of the succinic acid group ( $\text{C}_n\text{H}_{n-2}\text{O}_3\cdot\text{HO}$ ) namely, suberic, adipic, pimelic, and lipic acid, and, besides these, cenanthylic acid, but no oxalic acid. With fuming nitric acid it yields, on the other hand, according to Redtenbacher<sup>3</sup> almost all the acids of the first group ( $\text{C}_n\text{H}_{n-1}\text{O}_3\cdot\text{HO}$ ).

In the oily products of the dry distillation of oleic acid Schneider<sup>4</sup> found that the atoms of carbon were to those of hydrogen in the ratio of 6 : 5; and on treating these products with concentrated nitric acid, he obtained the same volatile acids which Redtenbacher obtained by the direct action of nitric acid on oleic acid.

*Composition.*—According to the above formula this acid contains:

Carbon,	.	.	.	.	.	36 atoms,	.	.	.	76.596
Hydrogen,	.	.	.	.	.	33 "	.	.	.	11.702
Oxygen,	.	.	.	.	.	8 "	.	.	.	8.511
Water,	.	.	.	.	.	1 "	.	.	.	3.191
										<hr/> 100.000

The atomic weight of the hypothetical anhydrous acid = 3412.5; its saturating capacity = 2.930.

*Combinations.*—The *oleates* are soft and greasy, and do not crystallize; like all the fatty acids, oleic acid has a strong tendency to form acid as well as basic salts. The neutral *oleate of lead* is a white powder which fuses at  $80^\circ$  into a yellow fluid, and is distinguished, by its solubility in boiling ether, from the lead-salts of all the solid fatty acids.

<sup>1</sup> Ann. d. Chim. et de Phys. T. 66, pp. 154–204.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 35, S. 86–108.

<sup>3</sup> Ibid. Bd. 59, S. 41–57.

<sup>4</sup> Ibid. Bd. 70, S. 107–121.

*Products of its Metamorphosis.*—Gottlieb,<sup>1</sup> who was the first to obtain pure oleic acid, and who, from his analyses, deduced the above formula, states that at an ordinary temperature, and when freely exposed to the atmosphere, this acid absorbs about 20 times its volume of oxygen, *without developing carbonic acid*. The thick fluid acid which is thus formed, and which now reddens litmus, contains 1 atom more of oxygen and 1 atom less of hydrogen than the pure oleic acid, being represented by the formula  $C_{36}H_{32}O_4.HO$ . This acid yields no *sebacic acid* on dry distillation. Hence it is that oleic acid, when not perfectly pure, that is to say, when changed by the access of oxygen, often yields only very little *sebacic acid*, while the quantities of capric and caprylic acids which are developed, remain constant.

If, however, oleic acid be exposed at a higher temperature to the action of oxygen, it rapidly assumes a rancid odor, becomes yellowish and more easily fusible, does not solidify so perfectly when exposed to cold, and its composition is represented by the formula  $C_{34}H_{33}O_5$ ; hence it may be regarded as a higher stage of oxidation of the radical of margaric acid than that obtained by Bromeis, and noticed in page 105.

*Elaidic acid* is, according to Gottlieb, perfectly isomeric with pure oleic acid, and is therefore represented by the formula  $C_{36}H_{33}O_3.HO$ . It is produced, as we have already mentioned, from oleic acid by the action of nitrous acid, without any development of gas; it crystallizes from an alcoholic solution, not in needles like oleic acid, but in large plates; it fuses at  $45^\circ$ , may be partially distilled undecomposed, dissolves readily in ether and alcohol, and strongly reddens litmus. On dry distillation elaidic acid yields no caprylic and capric acids, in which respect it differs essentially from oleic acid. In the fluid state this acid abstracts oxygen from the air, although less rapidly than oleic acid, and becomes converted, according to Gottlieb, into a higher stage of oxidation of the same radical, which we may assume to exist in oleic and elaidic acids, namely into  $(C_{36}H_{33})O_8$ . How the metamorphosis of oleic into elaidic acid exactly takes place, or on what it depends, are points on which as yet we have no certain knowledge.

*Preparation.*—This acid also is obtained by the saponification of vegetable and animal fats; the oleate of potash is extracted from the soap with cold absolute alcohol; the aqueous solution of oleate of potash is then precipitated with acetate of lead, and the oleate of lead (free from the margarate) is taken up from the dried precipitate by boiling ether. If the lead-salt, after the removal of the ether, be decomposed with carbonate of soda, and if the resulting soda-salt be decomposed with sulphuric acid, we obtain a somewhat brownish oleic acid mixed with products of oxidation. In order to obtain the acid in a state of perfect purity, we must, according to the directions of Gottlieb, treat it with an excess of ammonia, and precipitate it with chloride of barium: the baryta-salt is then to be repeatedly crystallized in moderately concentrated boiling alcohol, till it form a dazzling white flocculent powder, which must be decomposed with tartaric acid and thoroughly washed with water. Pure oleic acid may be more rapidly obtained by causing it to

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 37-67.

solidify by exposing it to a temperature of  $6^{\circ}$  or  $7^{\circ}$ , and then submitting it to strong pressure; as the above-mentioned products of oxidation of oleic acid remain fluid, they become absorbed in the filtering paper, and leave the oleic acid in a state of purity. Further, the water must only be removed while the oleic acid is exposed to a stream of carbonic acid, and all operations upon it should be conducted at a temperature below  $+10^{\circ}$ , since it very rapidly becomes decomposed.

*Tests.*—If it be required to test a fat or a mixture of fatty acids accurately for oleic acid, we must first isolate this acid by one of the methods which we have described, and obtain it in a state of at least tolerable purity, so as to enable us to ascertain the solubility of the lead-salt in hot ether. Moreover, oleic acid possesses the distinctive character of being the only one either of the oily or solid fatty acids which, on dry distillation, yields *sebacic acid*—an acid which may be distinguished from the simultaneously formed capric and caprylic acids by its crystallizability, and which we may easily separate from them and recognize, by forming and crystallizing its baryta-salt.

### *Physiological Relations.*

*Occurrence.*—Oleic acid, in combination with alkalies, exists in the blood and in the bile, and, in lesser quantity, in most of the other animal fluids, except the urine: in combination with oxide of lipyl, as a haloid salt, it occurs in the fat of the cellular tissue, and, indeed, wherever free fat is found in the animal body.

*Uses.*—As the vegetable fats are, for the most part, far richer in oleate of oxide of lipyl (olein) than animal fats, there seems to be a reason for the assumption that one of the uses of oleic acid in the animal body, is to form the more solid fats, margaric and stearic acids;—a view which is supported by the nature of the action of atmospheric air on oleic acid (to which we have already referred), and by its conversion into an acid having the radical of margaric acid. It might, however, have been expected *à priori* that animal fat would contain more margarate than oleate of oxide of lipyl, since oleic acid or an oleate is more rapidly consumed than margaric acid. We must, however, here, as in many other departments of physiological chemistry, rather abstain wholly from all conjectures than allow ourselves to be led astray by mere fancy. Let us rather wait for further facts to serve as substrata on which to establish a strictly logical hypothesis. Generally speaking, the function of oleic acid in the animal body coincides with that of the other fatty acids: but we shall return to this subject in a future part of this volume.

*Origin.*—In our remarks on the fats, we shall consider the question whether the animal body possesses the power of forming margaric and oleic acids as well as stearic acid.

### DOEGLIC ACID.— $C_{38}H_{35}O_3.HO$ .

This acid, which was discovered by Scharling<sup>1</sup> in the train oil of *Ba-*

<sup>1</sup> Journ. f. pr. Ch. Bd. 43, S. 257-271.



*laena rostrata*, is obtained from the lead-salt which is taken up by ether, precisely in accordance with Gottlieb's method of purifying oleic acid. At  $+16^{\circ}$  it is perfectly fluid, but solidifies at a few degrees above  $0^{\circ}$ : it is yellow and reddens litmus; on dry distillation it yields *no sebacic acid*. This acid is, moreover, not combined with oxide of lipyl in the Doegling train-oil (at least it yields no glycerine on saponification), but probably with *doeglic oxide*,  $C_{24}H_{25}O$ , a body similar to the ether-like haloid bases, whose existence and composition Scharling, however, only infers from the analysis of the unsaponified Doegling train-oil and the absence of glycerine.

#### DAMALURIC ACID.— $C_{14}H_{11}O_3.HO$ .

This acid, discovered by Städelér, was found together with damoleic acid amongst the products of distillation of cows' urine treated with hydrochloric acid; it is an oily fluid with a peculiar odor, not unlike that of valerianic acid, is somewhat heavier than water, in which it is slightly soluble, reddens litmus powerfully, and yields a white precipitate with basic acetate of lead, which under the microscope appears crystalline. Its silver-salt is not affected by light; its baryta-salt is crystallizable, soluble in water, renders turmeric paper brown, does not fuse when heated, and leaves, after smouldering, carbonate of baryta in the form of the original salt.

#### DAMOLEIC ACID.— $C_{26}H_{23}O_3$ .

This acid, also discovered by Städelér, occurs with damaluric acid amongst the volatile acids of cows' urine;<sup>1</sup> it is fluid, heavier than water, in which it is only slightly soluble, reddens litmus, and forms a crystallizable salt with baryta, which, however, fuses on the application of heat.<sup>2</sup>

### NON-NITROGENOUS RESINOUS ACIDS.

#### LITHOFELLIC ACID.— $C_{40}H_{36}O_7.HO$ .

##### *Chemical Relations.*

*Properties.*—This acid crystallizes in small, six-sided, right prisms, is readily pulverizable, fuses at  $205^{\circ}$ , and solidifies again in a crystalline form, if it has not been too highly heated; if, however, this has been the case, it solidifies into a vitreous, negatively idio-electric mass; in this condition it fuses at  $105^{\circ}$  to  $116^{\circ}$ ; by solution in, or mere moistening,<sup>3</sup> with alcohol, it returns to its former condition, being difficult to fuse again; when heated in the air, it volatilizes in white vapors with an aromatic odor; when inflamed it burns with a bright, smoky flame; it is decomposed by dry distillation; it is insoluble in water, dissolves readily in hot alcohol, but only slightly in ether; acetic acid dissolves

<sup>1</sup> Nachr. d. Ges. d. Wiss. z. Göttingen, 1850, S. 233–248.

<sup>2</sup> [The above notices of Damaluric and Damoleic acids have been introduced into the work by the translator, Dr. Day.—Am. Ed.]

it freely; acids precipitate it from its soluble salts as an amorphous coagulum.

*Composition.*—Ettling and Will,<sup>1</sup> from their analyses, calculated for it the formula  $C_{42}H_{36}O_8$ ; Wöhler,<sup>2</sup> from his analyses, deduced the formula  $C_{40}H_{36}O_8$ ; and Berzelius,<sup>3</sup> judging from the saturating capacity of the acid, considers the formula  $C_{40}H_{36}O_7.HO$  as the most correct: hence it must be regarded as containing:

Carbon,	.	.	.	.	.	40 atoms,	.	.	.	.	70.381
Hydrogen,	.	.	.	.	.	36 "	.	.	.	.	10.557
Oxygen,	.	.	.	.	.	7 "	.	.	.	.	16.422
Water,	.	.	.	.	.	1 "	.	.	.	.	2.640
											100.000

Hence the atomic weight of the hypothetical anhydrous acid (according to the above formula) = 4150, and its saturating capacity = 2.41.

*Combinations.*—This acid dissolves readily both in caustic ammonia and in carbonate of ammonia, but on evaporation of the solution it remains free from ammonia; the salts of baryta and lime throw down no precipitate from this solution: moreover, it dissolves readily in caustic potash, but is precipitated by an excess of potash as well as by hydrochlorate of ammonia; on the addition of the salts of lead or silver to a saturated potash-solution of this salt with only a faintly alkaline reaction, there is a white precipitate which, on warming, becomes plaster-like. Ettling and Will have obtained a silver-salt which crystallized in needles; Wöhler, however, only obtained an amorphous salt.

*Preparation.*—This acid, which was originally discovered by Göbel, is extracted from certain intestinal concretions by hot alcohol; the solution is decolorized by animal charcoal, and gradually evaporated.

*Tests.*—This acid may be recognized with tolerable certainty by the properties which we have already enumerated. If, however, it be found in other places than in intestinal concretions, it should always be submitted to an elementary analysis.

#### *Physiological Relations.*

*Occurrence.*—According to the researches of Merklein and Wöhler,<sup>5</sup> as well as those of Taylor,<sup>6</sup> this body exists only in certain bezoars, which are obtained from the intestines, and especially from the stomach of many species of goats inhabiting the East; other bezoars contain ellagic acid.<sup>7</sup>

*Origin.*—Whether lithofellic acid takes its origin in the bile, or is dependent on the use of resinous food, is as yet undecided, since its similarity to the resins is as great as to the resinous acids of the bile. Its analogy with ellagic acid certainly speaks in favor of its origin from the food; if, however, Taylor's view, that concretions containing lithofellic acid are frequently found in the stomach, be confirmed, it is obvious that they cannot owe their origin to the bile.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 39, S. 237-244.    <sup>2</sup> Pogg. Ann. Bd. 54, S. 255.

<sup>3</sup> Jahresber. Bd. 22, S. 560.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 39, S. 237.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 55, S. 129-143.

<sup>6</sup> Lond. Edinb. and Dubl. Phil. Mag. vol. 28, pp. 192-200.

<sup>7</sup> [Funke has only obtained it in prismatic forms, as represented in Fig. 3.]

CHOLIC ACID.— $C_{48}H_{39}O_9 \cdot HO$ .*Chemical Relations.*

*Properties.*—This acid crystallizes in tetrahedra, and more rarely in square octohedra (Fig. 3), is colorless, glistening, and easily pul-

Fig. 3.



Cholic Acid.

verized; the crystals effloresce on exposure to the air; the acid is bitter, leaving a faint sweetish after-taste; it is soluble in 750 parts of boiling, and in 4000 parts of cold water; it dissolves very readily in alcohol, especially when heated, and in 27 parts of ether. The acid, in crystallizing from ether, forms rhombic tablets, and in this form it contains 2 atoms of water, while from alcohol it crystallizes in tetrahedra with 5 atoms of water; the acid separated from alcohol by the addition of water contains 2 atoms of water, which it loses at  $100^\circ$ , while the tablets only lose 1 atom at that temperature. Moreover, this acid strongly reddens litmus, fuses at  $195^\circ$ , and at a higher temperature undergoes decomposition; above  $195^\circ$  it loses its atom of basic water, and is converted into choloidic acid, and at  $290^\circ$  it becomes converted into dyslysin (Strecker);<sup>1</sup> when inflamed it burns with a clear flame. It dissolves in sulphuric acid; and if to this solution we add a drop of syrup (1 part of sugar to 4 of water), the fluid assumes a beautiful purple-violet tint. If cholic acid be boiled for some time with hydrochloric acid it ceases to be crystallizable, and is converted into the resinous *choloidic acid*; and on further prolonging the boiling, the body, at the same time that it loses its solubility in alcohol and alkalies, also parts with its acid properties and then forms *dyslysin*. By the action of boiling nitric acid, it is for the most part converted into capric, caprylic, and cholesteric acids, without yielding oxalic acid or the volatile acids of the first group.

*Composition.*—This acid, which was first obtained in a state of purity by Demarçay, has been recently examined with much care by Strecker.<sup>2</sup>

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 375–378.<sup>2</sup> Ibid. Bd. 66, S. 1–61.

He found that it was constituted in accordance with the above formula. It consequently consists of:

Carbon, . . . . .	48 atoms,	70.588
Hydrogen, . . . . .	89 "	9.559
Oxygen, . . . . .	9 "	17.647
Water, . . . . .	1 "	2.206
		<hr/> 100.000

Consequently the atomic weight of the hypothetical anhydrous acid = 4987.5, and its saturating capacity = 2.005.

Mulder,<sup>1</sup> from his analyses of this acid, has deduced for it the formula,  $C_{50}H_{36}O_6 + 5HO$ .

Strecker, who by his admirable memoir on the bile of the ox, has done so much to advance our knowledge regarding this very obscure fluid, has unfortunately increased the existing confusion regarding cholic acid by giving it the new name of *cholalic acid*, while he applies the name of cholic acid to another acid which we shall subsequently describe. It is, however, true that Gmelin applied the term cholic acid to that acid of the bile in whose salts he recognized a sweet taste, and regarded it as a nitrogenous acid; but the non-nitrogenous acid first obtained in a state of purity by Demarçay, which in its mode of preparation and in its properties is identical with that which is here described, has so long been known as cholic acid that this name ought to be retained, and the more so because the new name of cholalic acid is by no means more expressive of its nature. We therefore retain the denomination which Demarçay, its discoverer, applied to it.

*Combinations.*—The *cholates* possess a bitter and at the same time a slightly sweet taste; they are all soluble in alcohol, but water dissolves only the alkaline cholates and cholate of baryta, and, to a very slight extent, cholate of lime. Cholic acid, with the aid of heat, expels the carbonic acid from solutions of the alkaline carbonates.

*Cholate of potash*,  $KO.C_{48}H_{39}O_9$ , is obtained in acicular crystals, by the evaporation of the alcoholic solution, or by the addition of ether to it. By spontaneous evaporation of the aqueous solution it forms a kind of varnish; the salt is insoluble in an excess of solution of potash, and, on the addition of caustic potash is precipitated in a gelatinous state. *Cholate of soda* and *cholate of ammonia* are very similar to it; the latter of these two salts loses the greater part of its ammonia on mere evaporation. *Cholate of lime*, when obtained by precipitation, is amorphous, but it crystallizes on the addition of ether. *Cholate of silver* is only very slightly soluble in water; it crystallizes, however, from a boiling solution.

*Products of its metamorphosis.*—*Choloidic acid*, as it exists in its salts, is perfectly isomeric with cholic acid; it is formed, as we have already mentioned, by boiling cholic acid with stronger acids. It may, however, be obtained by boiling together for some hours hydrochloric acid and that portion of the alcoholic extract of bile which is precipitable by ether; by solution in alcohol and precipitation by ether, it may be readily purified. It is a peculiarity of choloidic acid that in its isolated

<sup>1</sup> Unters. üb. d. Galle, übers. v. Völkel. Frank. a. M. 1847. S. 26.

state it contains no basic water, and may therefore be prepared in an actually anhydrous state; it forms a white, amorphous, resinous, pulverizable mass, which is insoluble in water, but dissolves freely in alcohol, and slightly in ether. The addition of water or ether to the alcoholic solution causes a milky appearance, and finally precipitates the acid in a resinous form; the alcoholic solution reddens litmus. When warmed, choloidic acid softens; at  $150^{\circ}$  it fuses, and at  $295^{\circ}$  it becomes converted into dyslysin, with the loss of 3 atoms of water. With concentrated sulphuric acid and sugar it gives the same reaction as cholic acid. When distilled with nitric acid, it yields not only the same volatile acids as oleic acid when similarly treated, but additionally choloidanic, cholesteric, and nitrocholic acids, and cholacrole (Redtenbacher).<sup>1</sup>

Its *salts* have a purely bitter taste, without any sweet after-taste; the acid is displaced from them by stronger acids, and even by carbonic acid, although, on the other hand, choloidic acid expels carbonic acid when heated with carbonates. The alkaline salts of this acid are soluble in water and in alcohol, but not in ether; they cannot be obtained in a crystalline state. *Choloidate of baryta*, although isomeric with the cholate, is not crystallizable, and is insoluble in water. With earths and metallic oxides this acid forms salts which are soluble in alcohol but insoluble in water.

*Dyslysin*  $C_{48}H_{36}O_6$  (Strecker),  $C_{50}H_{36}O_6$  (Mulder), is obtained from cholic or choloidic acid by one of the methods which we have already mentioned; the mass thus formed is extracted with water and alcohol, and dissolved in ether, from which it is again precipitated by alcohol; it is now of a grayish-white color, and the extent of its solubility depends upon the degree of its purity; it is, however, insoluble in acids and alkalies. When fused with hydrate of potash, or boiled with an alcoholic solution of potash, dyslysin is reconverted into choloidic acid.

From the choloidic acid of Demarçay, Berzelius has separated two acids, which he has named *fellic* and *cholinic acids*; <sup>2</sup> he, like Mulder, regards choloidic acid as an admixture of these two acids; it is to be regretted that Strecker, in his otherwise admirable investigation, has not made that reference to these substances which they deserve; for other chemists as well as Mulder may repeat the experiments and confirm the statements of Berzelius. We shall content ourselves in the present place, with indicating the most important points of difference between these two acids.

*Cholinic acid* ( $C_{50}H_{38}O_8$ , Mulder) forms white and bright flocculi, insoluble in water, and which, on drying, become brown and pulverizable. Its baryta and lead-salts have a tendency to cake together, and are *almost insoluble in alcohol*; the ammonia-salt of this acid separates as a white, saponaceous mass.

*Fellic acid* ( $C_{50}H_{40}O_{10}$ ) forms snow-white flocculi, which when dried become pulverizable; it is slightly soluble in water, and its solubility in ether is even less than that of cholinic acid. Its baryta and lead-salts are *soluble in alcohol*.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 145-170.

<sup>2</sup> [In the German these acids are termed *Fellinsäure* and *Cholinsäure*: we adopt the phrase *cholinic acid* for the latter word, as *cholic acid* is a pre-engaged name.—G. E. D.]

Redtenbacher distilled nitric acid over choloidic acid as long as vapors of nitrous acid continued to be developed, and he found in the receiver acetic, butyric, valerianic (?), caproic, cœnanthylic, caprylic, pelargonic, and capric acids (precisely the same as he obtained when oleic acid was similarly treated), and besides these, a heavy, stupifying oil, which, when treated with alkalis, was decomposed into *nitrocholic acid* and *cholacrole*; while in the retort there remained, as if proof against the further action of nitric acid, *oxalic*, *choloidanic*, and *cholesteric* acids.

*Cholacrole*,  $C_8H_5N_2O_{13}$ , is a yellow oil with a pungent, overpowering, cinnamon-like odor, dissolving readily in alcohol and ether, but difficult of solution in water; it is indifferent towards both acids and alkalis, and is decomposed at  $100^\circ$  with the development of nitrous acid, and sometimes with slight decrepitation.

*Nitrocholate of potash*,  $KO.C_2HN_4O_9$ , occurs in lemon-yellow, square tablets, has a faintly overpowering odor, decrepitates at  $100^\circ$ , is decomposed when boiled with water, and is not precipitated by metallic salts.

On pouring into a large test-glass the thick, brownish-yellow mass which remains in the retort, it separates on cooling into two layers, of which the upper is frothy, and consists of crystals of choloidanic acid, while the lower is of a yellowish-brown color, acid and bitter.

*Choloidanic acid*,  $C_{16}H_{12}O_7$ , crystallizes in satiny, hair-like prisms; when dry, it resembles asbestos; it is difficult of solution even in hot water, but dissolves freely in alcohol; it reddens litmus, and is decomposed at a high temperature, but is unaffected by hydrochloric or nitric acid. Its salts, even those of the alkalis, are insoluble or difficult of solution, and do not crystallize.

In this yellowish-brown mother-liquid there are also contained oxalic acid, a resinous mass, and cholesteric acid.

*Cholesteric acid*,  $C_8H_4O_4.HO$ , occurs as a light yellow mass, resembling cherry-gum; it has a well-marked acid and bitter taste, abstracts water from the air, dissolves both in water and in alcohol, the solution being of a yellow tint, and decomposes when heated; its compounds with alkalis and alkaline earths do not crystallize, and are soluble in water, but its compounds with metallic oxides are insoluble. The silver-salt dissolves in boiling water, from which it is deposited, on cooling, in crystalline incrustations.

*Preparation.*—Cholic acid, which occurs in the bile conjugated with nitrogenous bodies, is most readily obtained by boiling the resinous masses precipitated by ether from the alcoholic solution of the bile with a dilute solution of potash for twenty-four to thirty-six hours, till the potash-salt that has separated begins to crystallize. The potash-salt must then be dissolved in water and the acid removed from it by hydrochloric acid. By the addition of a few drops of ether, the acid which was previously resinous becomes crystalline, solid, and admits of trituration; it must be pulverized, washed with water, recrystallized in alcohol, and finally treated with a little ether in order to remove any coloring matter that may be attached to it.

*Tests.*—Cholic acid even when not perfectly pure may be recognized by its reaction with sugar and sulphuric acid. This reaction, which was first discovered by Pettenkofer,<sup>1</sup> occurs with no other substance than

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 90-96.

cholic acid; it is, however, perfectly immaterial whether the cholic acid be already metamorphosed into choloidic acid, or whether it be combined with its adjuncts, as a conjugated acid. Hence we can apply this admirable test to discover generally either the presence of bile or of one of its derivatives. The following is the best method of proceeding. The alcoholic extract of the fluid to be tested for biliary matter must be dissolved in a little water, with which we must then mix a drop of a solution of sugar (in the proportion of 1 part of sugar to 4 of water); and pure English sulphuric acid, free from sulphurous acid, must be added by drops to the mixture; the fluid now becomes turbid from the separation of the cholic acid, but on the gradual addition of sulphuric acid the turbidity disappears, and the fluid again becomes perfectly clear; for the first few moments its color is yellowish, it very soon however becomes of a pale cherry color, then of a deep carmine, of a purple, and finally, of an intense violet tint. As, indeed, in all experiments, some practice and attention to certain rules are requisite, without which we may easily fail to apply this test successfully to the detection of bile. For instance, we must avoid the addition of too much sugar, as this is a substance which is easily rendered brown or black by sulphuric acid; and we must be especially careful, as Pettenkoffer himself showed, while adding the concentrated sulphuric acid, not to allow the temperature much to exceed  $50^{\circ}$ ; but the reaction equally fails when we carry our caution too far, and attempt to avoid any elevation of temperature when the sulphuric acid is added; indeed, my own experience leads me to believe that an elevation of temperature nearly to  $50^{\circ}$  is requisite for the success of the experiment. Should the fluid at first assume only a cherry-red or a deep carmine tint, it must be allowed to stand for some time, after which the intense violet color becomes developed. It is, moreover, immaterial which kind of sugar is used for this test: acetic acid may also be employed in place of sugar.

Van den Broek<sup>1</sup> maintains that the reaction also takes place with mere biliary matter independently of the sugar, but I have never found this to be the case; without sugar the fluid has at most attained a red or reddish-brown tint, but never the characteristic, deep violet color. But although Van den Broek is wrong on this point, there are other reasons why his view is correct, that this reaction is inapplicable as a test for sugar; in the first place, because we have the same reaction when other bodies, as for instance, acetic acid, are substituted for sugar, and, secondly, because we have many better and more certain means of discovering this substance. F. Kunde<sup>2</sup> ascertained while working in my laboratory, that oleic acid, and likewise certain ethereal oils possess the property of producing the same color with concentrated sulphuric acid and a little sugar as cholic acid and its conjugated compounds; and Schultz<sup>3</sup> has independently arrived at the same result. I am not, however, inclined to believe from my own observations that there is much probability of any mistake arising from this circumstance, since olein and oleic acid when mixed with sulphuric acid and sugar only slowly gave rise to this coloration, the process being dependent on an absorption of oxygen, and,

<sup>1</sup> *Hollandische Beiträge*. Utrecht u. Düsseldorf. 1846. S. 100-102.

<sup>2</sup> *Dissert. inaug.* Berol. 1850.    <sup>3</sup> *Ann. d. Chem. u. Pharm.* Bd. 71, S. 266-277.

therefore only taking place in thin layers, as for instance, on a watch-glass.

If it should be necessary to separate the cholic acid from the conjugated biliary acids, or from choloidic acid, as is sometimes required in the examination of the blood, urine, and excrements, the best method is to acidulate the alcoholic extract with a little sulphuric acid, and to extract with ether, in which the conjugated biliary acids and choloidic acid are all but insoluble. As the cholate of baryta is soluble and crystallizable, which is not the case with the choloidate, we may thus as well as by the crystallizability of free cholic acid, readily distinguish between cholic and choloidic acids; the biliary acids are not only perfectly insoluble in ether, but one of them, when boiled with potash, yields ammonia, and the other, when similarly treated with hydrochloric acid, yields taurine, which, as we shall presently show, may be easily recognized under the microscope by the form of its crystals.

### *Physiological Relations.*

*Occurrence.*—In the *bile* we neither find cholic nor choloidic acid isolated from its respective adjunct; hence either within the animal body, in the gall-bladder, or after removal from the organism, it seems to have already passed into a state of decomposition, or else one of these acids must have been produced by the chemical treatment to which the bile has been subjected.

In examining the *blood* and the *urine* of patients suffering from diseases in which the liver is not directly implicated, we not unfrequently meet with substances yielding the above-described reaction for bile; I have, however, never satisfied myself in such cases, by any method, that either the one or the other of the biliary acids could be recognized with certainty. We shall treat more fully of the occurrence of these biliary matters in the blood and urine in our observations on the conjugated biliary acids. (See also “Blood” and “Urine.”)

In healthy *solid excrements* Pettenkofer<sup>1</sup> found no substance yielding this biliary reaction; the dejections in cases of diarrhoea, on the other hand, always contained a substance yielding this reaction. I have, however, always been able to detect a little cholic acid in perfectly normal excrements.

The alcoholic extract of previously dried solid excrement presented no reaction with sulphuric acid and sugar; but on further treating this extract with ether, and on purifying the residue of the ethereal solution, by means of water, from the fatty acids which are always mixed with it, I found that the somewhat concentrated aqueous solution (of this ethereal extract) presented the biliary reaction most beautifully. On using a larger quantity of material, the acid was obtained in a crystalline state; as it yielded no ammonia when treated with potash, and as its baryta-salt was soluble, it could hardly have been any other than cholic acid.

In the *intestinal canal* we can detect the presence of bile in the contents of the whole of the small intestine, by the addition of sulphuric acid to the alcoholic extract, in the manner above described.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 90-96.



If I rightly recollect, Pettenkofer informed me, in a private communication, that he had already made this observation. I have repeatedly convinced myself of its accuracy in animals; in the case of an intestinal fistula where it could not be determined with certainty whether the perforation was in the small or large intestine, and where no conclusion could be drawn from the absence of villi, the diagnosis was established by the bile-test. It was subsequently proved that the fistula occurred in the small intestine.

That substances containing or yielding cholic acid sometimes occur in *exudations*, requires no proof, as the blood is frequently overloaded with such matters.

I will here only mention that in the dropsical exudations occurring in a case of granular liver, and in another case of insufficiency of the mitral valves with stoppage of the biliary ducts, I found a considerable quantity of biliary matter. This subject is more fully noticed in the chapter on "Exudations."

The presence of biliary matters in morbid *saliva* and *expectoration*, is asserted by Wright,<sup>1</sup> but has not been proved.

*Origin.*—As we must return in a future page, to the different opinions which are maintained regarding the origin of the essential constituents of the bile, we shall here only notice such points as chemically elucidate the formation of cholic acid. That cholic and choloidic acids proceed from conjugated biliary acids, has been already mentioned; but according to the theoretical views which are at present maintained, cholic acid exists preformed in these biliary acids, just as in every conjugated acid we regard the true acidifying group of atoms as already formed. Without alluding here to the question whether the bile is primarily formed in the blood or in the cells of the liver, we will merely inquire what substances in the animal body yield that group of atoms which we call cholic acid? Even if many physiological and pathological facts did not support the view that the fats yield the principal material for the formation of the bile, the experiments of which we have made mention regarding the products of oxidation of cholic and choloidic acids would lead us to the belief that these bodies are closely allied to the fats, and especially to oleic acid; for we have seen that Redtenbacher has obtained from choloidic acid when treated with nitric acid precisely the same volatile acids (of the first group) as were yielded by oleic acid under similar treatment, independently of other specific substances. These latter may appropriately be regarded as arising from a group of atoms still hidden in the cholic acid, which group must be assumed to be an adjunct in the cholic acid. For if it be not improbable that such simple acids as acetic acid, butyric acid, &c., are to be regarded as conjugated acids, we are almost compelled to regard an acid like cholic acid with so high an atomic weight, and so considerable an amount of oxygen (that is to say, with so small a saturating capacity) as a conjugated acid.

From the circumstance of cholic acid yielding these products of decomposition, we may conjecture that it is a conjugated oleic acid; and assuming this to be the case, there remains as the adjunct the group of atoms ( $C_{48}H_{39}O_9 - C_{36}H_{33}O_3 =$ )  $C_{12}H_6O_6$  whose percentage composition is the

<sup>1</sup> The Lancet, 1842-3. Vol. 1, p. 559.

same as that of the cholesteric acid found by Redtenbacher in the products of decomposition of choloidic acid, and which is therefore polymeric with it (for  $C_{12}H_6O_6 : C_8H_4O_4 = 3 : 2$ ). That such polymeric groups of atoms frequently occur in the animal body as conjugated compounds, is obvious from Strecker's<sup>1</sup> discovery, that hippuric acid is, like the amides (see p. 44), decomposed into nitrogen, water, and an acid whose composition was found to be  $C_{18}H_8O_8$ , but which probably exists as a hydrate,  $C_{18}H_{10}O_9$ , and in that case is polymeric with cholesteric acid. That cholic acid is oleic acid conjugated with the atomic group  $C_{12}H_6O_6$  is merely a hypothetical view which, founded on certain chemical facts, may seem to indicate a direction for future experimental investigations, but cannot warrant us in advancing further in this domain of the imagination. We postpone for the present entering into the consideration of other hypotheses tending to elucidate the origin of the group of atoms conjugated with oleic acid.

We must necessarily defer our remarks on the possible use of cholic acid in the animal body, till we treat of the uses of the conjugated cholic acids and of the bile generally.

## NITROGENOUS BASIC BODIES.

SUBSTANCES of this nature occur principally in the vegetable kingdom; those requiring a notice in animal chemistry are almost all only artificial products of known animal matters: in as far, however, as they, like many of the acids which have been already described, throw much light on the constitution of the bodies from which they are derived, they must not be passed over in a work of this nature. As there exists no true alkaloid without nitrogen, the basicity of this class of bodies may be regarded as essentially depending on the amount of nitrogen which they contain; and in further confirmation of this view, we may bring forward the fact that the saturating power of these bodies is perfectly independent of the amount of oxygen which they contain. Indeed it rather depends in most cases on the amount of nitrogen; that is to say, 1 equivalent of the nitrogen of the base requires 1 equivalent of acid in order to form a neutral salt. Berzelius has, therefore, advanced the opinion that the nitrogenous bases are merely ammonia-compounds, with either a non-nitrogenous or a nitrogenous body as an adjunct. The principal argument in favor of this view is, that these bases, like pure free ammonia, cannot unite with oxygen acids, without simultaneously assimilating an atom of water, but that, on the other hand, they combine with hydrochloric and other hydrogen acids, without a separation of water; finally, they resemble ammonia in this respect, that the combination of their hydrochlorates with bichloride of platinum, are, like ammonio-chloride of platinum, difficult of solution. Moreover, that the nitrogen is not the direct cause of the basicity seems probable, from the circumstance that the saturating power of the substance, even when it contains several

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 52 ff.

equivalents of nitrogen, for the most part corresponds with only one equivalent; so that only this one equivalent is to be regarded as pertaining to the ammonia, and the remainder of the nitrogen to the adjunct.

These organic bases are divisible into two tolerably well-marked groups, according as they contain or are devoid of oxygen: as the former are, without exception, volatile, and the latter not so, we might also class them as volatile and non-volatile bases.

#### NON-OXYGENOUS ALKALOIDS.

The bodies of this group are very similar in their empirical composition to the nitriles which we have already described: in their rational composition there can, however, be no similarity, as they are essentially different in their chemical properties. The nitriles never show any basic properties, while the alkaloids cannot be decomposed into oxygen acids and ammonia either by acids or by alkalies, nor with potassium do they form cyanide of potassium. If, therefore, Berzelius's view, that the alkaloids are conjugated ammonia, find a confirmation in any substances, it must be in the non-oxygenous alkaloids, which in all their combining relations present so many analogies with ammonia that we might regard it as the representative of this group. Even the mode of preparing certain alkaloids, as, for instance, thiosinamine, affords evidence in favor of this view of the subject.

It is well known that, on treating cyanic acid with potash, there is a development of ammonia ( $\text{C}_2\text{NO} \cdot \text{HO} + 2\text{HIO} + 2\text{KO} = 2\text{KO} \cdot \text{CO}_2 + \text{H}_3\text{N}$ ); on heating cyanate of oxide of methyl or cyanate of oxide of ethyl with potash, a strongly basic alkaloid, similar to ammonia, is produced; here we feel almost compelled to assume that ammonia is formed from the cyanic acid just as from the free acid, and that this ammonia is conjugated with the carbo-hydrogen of the methyl or the ethyl ( $\text{C}_2\text{H}_2$  or  $\text{C}_4\text{H}$ ), and thus produces the alkaloid.

Urea presents perfectly similar reactions; when treated with alkalies it develops ammonia; and Wurtz<sup>1</sup> has shown that these alkaloids may be prepared in such a manner that acetate of urea, when heated with potash, shall yield the same alkaloid as is obtained by the action of potash on cyanate of oxide of methyl, namely  $\text{C}_2\text{H}_5\text{N}$ , while metacetate of urea, similarly treated, gives the same alkaloid as is obtained by the action of potash on cyanate of oxide of ethyl, namely  $\text{C}_4\text{H}_7\text{N}$ . Although these substances may either be regarded as pertaining to the class of ethers in which the oxygen is replaced by amide,  $\text{C}_4\text{H}_5\text{O} \sim \text{C}_4\text{H}_5\text{H}_2\text{N}$ , or as ammonia in which the third atom of hydrogen is replaced by methyl or ethyl, the most simple and probable explanation seems to be, that they should be regarded as conjugated ammonia-compounds  $= \text{C}_2\text{H}_2 \cdot \text{H}_3\text{N}$ , and  $\text{C}_4\text{H}_4 \cdot \text{H}_3\text{N}$ .

As was already mentioned, we shall here only notice those alkaloids which may be obtained from the decomposition of certain animal matters.

Many of these volatile alkaloids are liquid, like the nitriles, but most of them are crystallizable. They have generally a nauseous odor and an acrid burning taste, are slightly soluble or altogether insoluble in water,

<sup>1</sup> Compt. rend. T. 38, pp. 223-227.

dissolve readily in alcohol, are most soluble in ether and in fatty and volatile oils, and react on vegetable colors. Their salts are, for the most part, crystallizable and readily soluble; but their combinations with bichloride of platinum are nearly or entirely insoluble.

### ANILINE.— $C_{12}H_7N$ .

#### *Chemical Relations.*

*Properties.*—This alkaloid forms a colorless, strongly refracting, oily fluid, with an aromatic odor; its specific gravity = 1.020, it remains fluid at  $-20^\circ$ , evaporates very rapidly at an ordinary temperature, begins to boil at  $182^\circ$ , dissolves slightly in water, and in every preparation in alcohol and ether, coagulates albumen, dissolves phosphorus and sulphur, and colors Dahlia (Georgina) paper green; when exposed to the air it becomes yellow, and is converted into a resinous mass; a solution of hypochlorite of lime, on the addition of a few drops, assumes a violet color; with nitric acid, on the other hand, aniline yields an indigo color, and, by prolonged action, is converted into picric acid; with dilute chromic acid it yields a black or greenish-blue precipitate.

*Composition.*—According to the above formula aniline contains:

Carbon, . . . . .	12 atoms, . . .	77.419
Hydrogen, . . . . .	7 " . . .	7.527
Nitrogen, . . . . .	1 " . . .	15.054
		<hr/> 100.000

Its atomic weight = 1162.5. According to Berzelius, aniline consists of ammonia conjugated with a carbo-hydrogen =  $C_{12}H_4$ .

*Combinations.*—Aniline forms very characteristic, and, for the most part, crystallizable salts, both with the oxygen and the hydrogen acids; in the former, but not in the latter case, the salts assimilating an atom of water.

The analogy between aniline and ammonia is further shown by the circumstance that it, like the latter, under certain conditions, may lose a portion of its hydrogen, and be converted with an acid deprived of a portion of its oxygen (and therefore with the formation of water) into combinations analogous to the amides, to which the term *anilides* has been applied (Gerhardt.)<sup>1</sup>

As the elements of cyanate of ammonia, immediately after they are brought together, group themselves in a different manner and form urea, so cyanic acid and aniline do not form a simple salt, but a body, from which neither aniline nor cyanic acid can be again obtained, namely, aniline-urea,  $C_{14}H_8N_2O_2$  (Hofmann.)<sup>2</sup>

Aniline may so assimilate cyanogen that the latter may be regarded as an adjunct, the newly-formed body, cyaniline, entirely retaining its basic properties (Hofmann.)<sup>3</sup>

Aniline probably affords stronger evidence than any other body yet examined in reference to this point, in favor of the substitution theory,

<sup>1</sup> Journ. de Pharm. et de Chim. 1845, Juill. pp. 53-56.

<sup>2</sup> Quart. Journ. of the Chem. Soc. of Lond. 1848. Vol. i. pp. 159-174.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 247 ff.

since not merely one, but several of its equivalents of hydrogen, may be replaced by chlorine, bromine, iodine, or hyponitric acid, without the group of atoms entirely losing its basic properties. (Hofmann,<sup>1</sup> and Hofmann and Muspratt.)<sup>2</sup> Finally, a base has been discovered in which aniline is combined with the adjunct *cyanilide*,  $C_{12}(H_6Cy)N$ ; to this the name of *melaniline* has been applied (Hofmann)<sup>3</sup>.

*Preparation.*—This body very frequently occurs as a product of the decomposition of nitrogenous matters; thus, for instance, it is found among the products of the dry distillation of animal substances, as bone-oil (Anderson).<sup>4</sup> As it had previously been obtained in various ways, it received several different names, as *cyanol*, *benzidame*, and *crystalline*, before its identity was fully established. It is most easily obtained in a state of purity by heating anthranilic acid ( $C_{14}H_6NO_3 + HO = 2CO_2 + C_{12}H_7N$ ), or phenate of ammonia ( $H_4NO.C_{12}H_5O = 2HO + C_{12}H_7N$ ), or from nitrobenzide and sulphuretted hydrogen ( $C_{12}H_5NO_4 + 6HS = 6S + 4HO + C_{12}H_7N$ ).

*Tests.*—We have already pointed out the manner in which aniline reacts with hypochlorite of lime, and nitric and chromic acids; by these tests we can easily recognize it even when it is not exhibited in a perfectly pure state.

#### *Physiological Relations.*

It is remarkable that this substance, which affects the organism so unpleasantly from its smell and taste, should, according to Wöhler and Frerich's experiments,<sup>5</sup> be free from all poisonous action.

#### PICOLINE.— $C_{12}H_7N$ .

*Properties.*—This body, which was formerly called *pyrrol*, is also a thin fluid, having a penetrating, rank, aromatic odor, and a burning, bitter taste; it remains fluid at  $-20^\circ$ , evaporates at an ordinary temperature, boils at  $133^\circ$ , and its specific gravity = 0.955; it turns red litmus blue, does not change on exposure to the atmosphere, and does not coagulate albumen. It is not colored by chloride of lime, and experiences no alteration from chromic acid.

Its *Composition* resembles that of aniline.

*Combinations.*—With acids it forms bitter-tasting salts, soluble in water and alcohol, and partially deliquescent, although not so easily crystallized as those of the aniline, and less readily changed by the action of the air.

*Preparation.*—This body was first discovered in coal-tar, and subsequently in the products of the distillation of bones from which the fat has been removed (Anderson).<sup>6</sup> It is obtained by fractional distillation.

This body is isomeric, or rather identical with the *aniline* or *benzidine* =  $C_{12}H_7N$  (see p. 81) obtained from nitrobenzide by ammonia and sul-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 40-57.

<sup>2</sup> Ibid. Bd. 57, S. 201-224.

<sup>3</sup> Ibid. Bd. 67, S. 61-78, and Bd. 68, S. 129-174.

<sup>4</sup> Phil. Mag. 3 Ser. vol. 33, p. 185.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 340.

<sup>6</sup> Phil. Mag. 3 Ser. vol. 33, pp. 174-186.

phuretted hydrogen; this benzidine must not be confounded with the *benzidine* =  $C_{12}H_6N$  (see p. 82), which was obtained by Zinin,<sup>1</sup> from azobenzide, ammonia, and sulphuretted hydrogen.

### PETININE.— $C_8H_{10}N$ .

*Properties.*—This alkaloid is a colorless, highly refracting fluid, having a sharp pungent odor and taste; it boils at  $79^\circ$ , is easily soluble in water, alcohol, and ether, gives a blue tint to red litmus, is the strongest base of all these alkaloids, and is not colored but decomposed by chloride of lime.

*Composition.*—According to the above formula it consists of:

Carbon,	.	.	.	.	.	8 atoms,	.	.	.	66.666
Hydrogen,	.	.	.	.	.	10 "	.	.	.	13.890
Nitrogen,	.	.	.	.	.	1 "	.	.	.	19.444
										<hr/> 100.00

Its atomic weight is = 900.0. According to Berzelius, the theoretical formula of this body would be =  $H_3N.C_8H_7$ .

*Combinations.*—The compounds of petinine with acids are readily crystallizable, unaffected by the atmosphere, and soluble in water and alcohol. Chloride of platinum and petinine,  $P.HCl.PtCl_2$ , forms golden-yellow crystals resembling iodide of lead, pretty soluble in cold water.

*Preparation.*—This base is the most volatile of those yielded by the dry distillation of gelatinous tissues. It is obtained from the mixture of basic bodies and ammonia by fractional distillation.

### ALKALOIDS CONTAINING OXYGEN.

Few substances of this group belong to zoo-chemistry; but they are more important in reference to physiological chemistry than the non-oxygenous alkaloids which we have just considered, as they have either been found preformed in the animal body, or are able to throw considerable light on the constitution of the substances yielding them, and on organic chemistry generally. We shall therefore only consider in any detail the following substances, viz.:—creatine, creatinine, tyrosine, leucine, sarcosine, glycine (glycocoll), urea, guanine, xanthine, taurine, and cystine; and here it will be necessary to obtain some acquaintance with the general chemical relations of all these bodies before we enter upon the consideration of each individually.

The oxygenous alkaloids do not yield in respect to their basicity to those containing no oxygen; for many of these bodies not only separate the oxides of the heavy metals from their salts but also liberate ammonia. Their basicity, however, exhibits such gradual differences that no accurate line of demarcation can be drawn between decidedly basic and indifferent nitrogenous bodies. Thus leucine and creatine are

<sup>1</sup> Journ. f. pr. Ch. Bd. 35, S. 93.

perfectly indifferent bodies, while sarcosine, which is homologous to leucine, and creatinine, which is so similar to creatine, are strongly basic; but as these indifferent bodies present a close theoretical relation to the basic bodies, or actually possess weak basic properties, we do not think that it is expedient to separate them.

There is no direct ratio between the saturating capacity of these bodies and the quantity of oxygen or even of nitrogen that they contain, for in creatinine, for instance, only the third part of the nitrogen contained in the body corresponds to the saturating capacity, while in xanthine it is the fourth, and in guanine only the fifth part. In these bodies the nitrogen may be similarly incorporated with other elements as an adjunct of the base; thus we have seen that nitrogen may be artificially added to aniline under the form of cyanogen or hyponitric acid, and that harmaline (from *Peganum harmala*) takes up hydrocyanic acid without changing its saturating capacity.

The greater number of the alkaloids containing oxygen are crystallizable; none are fluid at an ordinary temperature; the majority have a more or less bitter taste: not being volatile, they have no odor; all are soluble in alcohol, a few in water, and none that we have here considered, in ether; although most alkaloids act on vegetable colors, none of those under consideration, excepting creatinine and sarcosine, exhibit this property.

Their salts are almost universally crystallizable and soluble in water as well as in alcohol; with bichloride of platinum their hydrochlorates form compounds which are either insoluble or difficult of solution; their oxygen salts cannot exist without 1 equivalent of water. The most strongly basic alkaloids are precipitated by tannic acid from dilute aqueous solutions.

Although many of the substances which we shall have to consider in this group do not possess any basic properties, and therefore do not, strictly speaking, belong to it, we have arranged them together, partly on account of the analogy exhibited in their empirical composition, and partly, because in a physiological point of view, they exhibit tolerably equal values, that is to say, they are derivatives of nitrogenous tissues. The bodies which we shall now consider, are:

Creatine,	.	.	.	.	.	.	.	.	$C_8H_9N_3O_4$ .
Creatinine,	.	.	.	.	.	.	.	.	$C_8H_7N_3O_2$ .
Tyrosine,	.	.	.	.	.	.	.	.	$C_{16}H_9N_3O_5$ .
Leucine, .	.	.	.	.	.	.	.	.	$C_{12}H_{11}N_2O_4$ .
Sarcosine,	.	.	.	.	.	.	.	.	$C_6H_7N_2O_4$ .
Glycine (Glycocoll),	.	.	.	.	.	.	.	.	$C_2H_5N_2O_4$ .
Urea,	.	.	.	.	.	.	.	.	$C_2H_4N_2O_2$ .
Xanthine,	.	.	.	.	.	.	.	.	$C_5H_4N_2O_2$ .
Guanine,	.	.	.	.	.	.	.	.	$C_{10}H_5N_5O_2$ .
Allantoine,	.	.	.	.	.	.	.	.	$C_8H_5N_4O_5$ .
Cystine, .	.	.	.	.	.	.	.	.	$C_6H_6NS_2O_4$ .
Taurine, .	.	.	.	.	.	.	.	.	$C_4H_7NS_2O_6$ .

#### CREATINE.— $C_8H_9N_3O_4$ .

##### *Chemical Relations.*

*Properties.*—This body forms transparent, very brilliant crystals,

belonging to the clinorhombic system and containing 2 atoms of water of crystallization; it is of a bitter, strongly pungent taste, and irritates

Fig. 4.



Creatine crystallized from hot water.

the pharynx; it loses its 2 atoms of water at  $100^{\circ}$ , and at a higher temperature becomes decomposed; it dissolves in 74.4 parts of cold water, and in boiling water in such quantity that, on cooling, the solution becomes consolidated into a mass of delicate glistening needles; it does not dissolve in less than 9410 parts of alcohol, and not at all in ether; it does not act on vegetable colors, and forms no definite salts with acids. It dissolves in baryta-water without undergoing any change, but when boiled with it, it becomes decomposed into ammonia and carbonic acid or into *urea* and *sarcosine*. It also dissolves unchanged in dilute acids; but when heated with strong acids, it becomes converted into *creatinine*, giving off 2 atoms of water.

*Composition*.—This body has recently been most carefully examined by Liebig;<sup>1</sup> from whose analyses the above formula is derived, and from which we find creatine to consist of:

Carbon,	.	.	.	.	.	8 atoms,	.	.	.	.	36.64
Hydrogen,	.	.	.	.	.	9 "	.	.	.	.	6.87
Nitrogen,	.	.	.	.	.	3 "	.	.	.	.	32.06
Oxygen,	.	.	.	.	.	4 "	.	.	.	.	24.43
											100.000

The 2 equivalents of water correspond to 12.08% of crystallized creatine. The atomic weight of the anhydrous substance is = 1637.5. Notwithstanding the various modes of decomposing creatine, no probable hypothesis can be adduced regarding its theoretical constitution. As it is almost wholly deficient in basic properties, it can hardly be regarded, according to Berzelius's view, as a conjugated ammonia; for it would in that case stand as  $\text{H}_3\text{N} \cdot \text{C}_8\text{H}_6\text{N}_2\text{O}_4$ , by which the deficient basic character is made more conspicuous; while Liebig's view of regarding crystallized creatine as a combination of ammonia and 2 equivalents of glycine (glycocoll), ( $\text{C}_8\text{H}_{11}\text{N}_3\text{O}_6 = \text{H}_3\text{N} + \text{C}_8\text{H}_8\text{N}_2\text{O}_6$ ), is opposed both by the con-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 257-290.



stitution of anhydrous creatine and by the deficiency in basicity. The decomposition of creatine by baryta-water into urea and sarcosine might indeed indicate that these bodies are its proximate constituents (for  $C_2H_4N_2O_2 + C_6H_7NO_4 = C_8H_{11}N_3O_6$ ), but this is not probable; for although we know that water is expelled on the union of two organic substances, we can no more assume that urea and sarcosine are present in the dry substance, than we could maintain that oxalic acid and ammonia are contained in oxamide, or valerianic acid and ammonia in valeronitrile.

*Preparation.*—Creatine is obtained, according to Liebig, from finely chopped *flesh*, that has been well kneaded with water and the fluid removed by pressure. The coagulable matters are then removed by boiling, from the fluid which is thus obtained, and the prosphates by caustic baryta; during the evaporation of the fluid filtered from these precipitates the surface will be continually covered with a membranous coating, which must from time to time be removed; after the fluid has been evaporated to  $\frac{1}{20}$ th of its volume it must be left to stand for some time, when the creatine will separate in needles. The crystals, when separated from the mother-liquid by filtering paper, must be washed with water and spirit of wine, and then again suffered to crystallize from hot water.

The following method is likewise given by Liebig for obtaining creatine from *urine*. The urine, after being treated with lime-water and chloride of calcium, and being filtered, is evaporated, and the greater part of the salts removed by crystallization; the mother-liquid poured off from the crystals is then decomposed with  $\frac{1}{4}$ th of its weight of a syrupy solution of chloride of zinc; after some days roundish granules of a compound of chloride of zinc and creatinine, with which some creatine is mixed, become separated; these granules, after being dissolved in boiling water, are treated with hydrated oxide of lead until there is an alkaline reaction. The fluid, after the removal of the oxide of zinc and chloride of lead by filtration, is freed from the lead and coloring matter by means of animal charcoal, and evaporated to dryness. The residue, consisting of creatine and creatinine, is treated with boiling alcohol, in which the latter dissolves readily, while the former is almost insoluble in it; by this means the two bodies can therefore be easily separated.

*Tests.*—In order to examine whether creatine be present in a fluid, (for which purpose a large amount of material is required), one of the above methods should be adopted, and the properties of any creatine-like substance compared with those of pure creatine. As, however, the determination of the atomic weight is not so readily made as in the acids, an elementary analysis is indispensable for the attainment of perfect certainty.

### *Physiological Relations.*

*Occurrence.*—Chevreul long since drew attention to this substance as a constituent of the decoction of flesh, but its presence was not again detected by any of the analysts who sought for it, until Schlossberger<sup>1</sup> found it in the *muscular tissue* of an alligator, and Heintz<sup>2</sup> proved its

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 49, S. 341.

<sup>2</sup> Pogg. Ann. Bd. 70, S. 476-480.

existence in beef, and was at the same time the first observer who accurately determined the composition of this body. Liebig may, however, be regarded as the first who made us thoroughly acquainted with it by his conclusive investigations regarding its chemical relations and the various situations in which it occurs. Liebig has examined so many different kinds of flesh for creatine, and so universally discovered it, that scarcely a doubt can now be entertained that creatine forms a constituent of the muscles of all the higher classes of animals. The quantity of creatine found in muscle is, however, exceedingly small. Liebig obtained only 36 grammes (consequently only 0.072%) of creatine from 100 pounds of lean horse-flesh; 30 grammes (or 0.07%) from 56 pounds of beef; but 72 grammes ( $= 0.32\%$ ) from 47 pounds of the flesh of lean fowls; consequently for every 100 parts of flesh there were only 0.07 or at most 0.32 parts of creatine, or 1 part of creatine to 1400 parts of flesh. Liebig has further convinced himself that lean flesh contains more creatine than fat flesh; and this may probably be the cause of proportionally a large quantity of creatine being found in the tissue of the heart of the ox.

I have likewise found creatine in the smooth muscles—in those namely of the stomach of the pig; and Siegmund<sup>2</sup> has subsequently detected it in the muscular substance of a pregnant uterus.

Verdeil and Marcet<sup>3</sup> have found both creatine and creatinine in the blood.

Liebig obtained the largest quantity of creatine from the flesh of fowls and martens; the quantity diminished progressively in the flesh of horses, foxes, roes, stags, hares, oxen, sheep, pigs, calves, and fishes. Liebig could frequently obtain only traces of creatine from fat flesh.

Gregory<sup>4</sup> has examined several kinds of flesh, according to Liebig's method, in reference to their amount of creatine. He found in 100 parts of bullock's heart from 0.1375 to 0.1418 parts of creatine, in the flesh of the cod-fish (*Gadus morrhua*) from 0.0935 to 0.17 parts, in the flesh of pigeons 0.0825 parts, and in the flesh of the skate (*Raja batia*) 0.0607 parts. Gregory especially recommends the flesh of the cod-fish, partly because it contains a proportionally large quantity of creatine, and partly because it most readily yields a pure, finely crystallized creatine. Sea-fish appear to contain much more creatine than fresh-water fish.

Schlossberger<sup>5</sup> has shown by direct experiment that human flesh presents no exception to the rule; 6 pounds of human flesh yielding about 2 grammes of creatine (therefore  $= 0.067\%$ ).

No creatine could be found in the substance of the *brain, liver, or kidneys*.

Creatine, together with creatinine, was first separated from the *urine* in the chloride of zinc compound by Heintz<sup>6</sup> and Pettenkofer<sup>7</sup> although they did not recognize its nature; Heintz<sup>8</sup> subsequently obtained pure

<sup>1</sup> Walther, Diss. inaug. med. Lips. 1851, p. 8.

<sup>2</sup> Verd. d. Phys.-med. Ges. zu Wurtzburg, Bd. 8, S. 50.

<sup>3</sup> Jour. de Chim. et de Phys. 8 Ser. T. 20, p. 91-93.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 64, S. 100-108.

<sup>5</sup> Arch. f. phys. Heilk. Bd. 7, S. 209-211.

<sup>6</sup> Pogg. Ann. Bd. 62, S. 602-606.

<sup>7</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 97-100.

<sup>8</sup> Pogg. Ann. Bd. 62, S. 602.

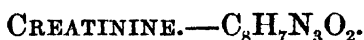
creatine from the zinc compound, and employed this substance for his analysis. Liebig, however, showed that the chloride of zinc compound, as yielded by urine, contained for the most part creatinine in chemical combination, the creatine being only mixed with it.

*Origin.*—When we remember that creatine occurs in the decoction of flesh, and is a highly nitrogenous body, we might be led to regard it as an important nutritive agent, and as taking an active part in progressive metamorphosis. The analogy which, in its chemical relation, and in its constitution, it presents to caffeine, might moreover tend to mislead those who class that substance among nutrient bodies, from its occurrence in certain kinds of food and in certain stimulants. But this analogy is here of very little moment, for we cannot place caffeine among the nutritive agents without giving a very great latitude to the term. A substance, of which a quantity from 2 to 10 grains will produce the most violent excitement of the vascular and nervous systems—palpitation of the heart, extraordinary frequency, irregularity, and often intermission of the pulse, oppression of the chest, pains in the head, confusion of the senses, singing in the ears, scintillations before the eyes, sleeplessness, erections, and delirium,—can scarcely be reckoned among articles of nutrition even by the homœopathist, and certainly not by physiologists, when they learn how quickly caffeine becomes decomposed in the organism, and gives rise to an increased secretion of urea.

The above-named results were yielded by experiments instituted on myself and several of my pupils with pure caffeine. Five persons (one of whom was Professor Buchheim, now at Dorpat), after taking from 5 to 10 grains of this substance, were unfit for any business during the next day, while, in an experiment which I formerly made on myself, 10 grains scarcely produced any perceptible\*action. In all the cases there was found to be augmentation of the total amount of urea excreted in twenty-four hours.

If, however, the analogy between creatine and caffeine does not demonstrate the nutrient qualities of the former, it must be asked, whether its occurrence in a substance so nourishing as the decoction of flesh, and its large amount of nitrogen, afford more conclusive evidence in this respect? With reference to the latter it may be assumed, that nature would not suffer substances even more highly nitrogenized than creatine, as the creatinine discovered by Liebig in the urine and the urea, to escape through the kidneys, if they could be employed to further advantage in the organism; since we find so careful a providence over recognized nutrient matters, as for instance albumen, &c., that even in disease they are only rarely found to escape with the excreta. The occurrence of creatine in the decoction of flesh affords even less evidence of its nutrient powers; for when we consider the small quantity in which it occurs in flesh, and the truly homœopathic nature of the dose which we take with the meat and broth we eat, we must regard its simultaneous appearance in the urine as a proof that its properties are not very highly esteemed in the organism; since, if they were so, this substance would probably not be discharged from the kidneys, but be retained in the same manner as albumen and gelatin. We think, however, that Liebig's complete chemical investigations of creatine, which were conducted in a manner worthy of so great a chemist, constrain us, even if unsupported by physiological proof,

to regard creatine as a *product of excretion*. From its chemical qualities, we regard creatine as a member of the series indicating the regressive metamorphosis from the point of the highest atomic weights to bodies of the simplest composition. The readiness with which creatine becomes decomposed into creatinine, urea, and sarcosine, which is isomeric with lactimide, all of which are undoubtedly products of excretion, proves beyond a doubt that creatine approximates more nearly to these substances than to albumen and fibrin, and indicates the great probability of creatine being decomposed even in the living body into these and other similar substances. Although such bodies as lactic acid, &c., may be employed for special purposes in the animal organism, they cannot, strictly speaking, be regarded as nutrient substances—that is to say, as materials for the renovation of nitrogenous tissues; and it is only in this light, and not in that of a supporter of heat, that we must consider creatine. Creatine is, however, a substance of the highest importance in relation to physiological chemistry; as it affords us a glimpse at the ever-recurring chemical changes which are associated with the functions of organs, and of which we have at present so little general knowledge.



*Chemical Relations.*

*Properties.*—This alkaloid forms colorless, very glistening crystals, belonging to the monoclinometric system; has almost as burning a taste

Fig. 5.



Creatinine crystallized from hot water.

as caustic ammonia, dissolves in 11.5 parts of water at an ordinary temperature, but more readily in hot water; while it requires about 100 parts of cold spirit to dissolve 1 part of creatinine, it is so freely soluble in hot spirit, that, on cooling, it again separates in crystalline masses; it is also slightly soluble in ether; it shows a strong alkaline action on vegetable colors, and it even separates ammonia from its salts. A moderately concentrated solution of nitrate of silver added to a solution of creatinine, causes a coagulation into a network of acicular crystals,

which dissolve on being boiled with water, and again appear when it cools. A solution of corrosive sublimate yields a curdy precipitate, which soon becomes crystalline; chloride of zinc likewise forms a crystalline granular precipitate. Bichloride of platinum, however, yields no precipitate when the solution is somewhat dilute.

*Composition.*—We are indebted solely to Liebig<sup>1</sup> for our knowledge of the composition of this substance. From the analyses of its salts he deduced the above formula, according to which it consists of:

Carbon, . . . . .	8 atoms, . . . . .	42.48
Hydrogen, . . . . .	7 “ . . . . .	6.19
Nitrogen, . . . . .	3 “ . . . . .	37.17
Oxygen, . . . . .	2 “ . . . . .	14.16
		<hr/>
		100.00

Its atomic weight = 1412.5. As this body possesses such strong basic properties, we may accept the hypothesis of Berzelius regarding its theoretical composition as the most probable one, namely, that it is ammonia conjugated with a highly nitrogenous body, containing exactly 1 atom less of hydrogen than caffeine =  $\text{H}_3\text{N} \cdot \text{C}_8\text{H}_4\text{N}_2\text{O}_2$ . Moreover, a comparison of the formulæ shows that creatinine contains exactly 2 atoms of water less than anhydrous creatine.

*Combinations.*—The combinations of creatinine with acids are, as far as is yet known, soluble in water and readily crystallizable.

*Hydrochlorate of creatinine*,  $\text{K} \cdot \text{HCl}$ , crystallizes from hot alcohol in short transparent prisms; from water, in broad leaves; with bichloride of platinum it yields an easily soluble compound which crystallizes in crimson prisms =  $\text{K} \cdot \text{HCl} + \text{PtCl}_2$ .

*Sulphate of creatinine*,  $\text{K} \cdot \text{HO} \cdot \text{SO}_3$ , forms concentrically grouped, transparent, square tablets, which lose no water at  $100^\circ$ , and remain perfectly translucent.

With the above-named *metallic salts* creatinine yields crystallizable compounds, all of which are basic double salts; with the salts of the oxide of copper it forms crystallizable double salts of a beautiful blue color.

*Preparation.*—The most simple method of obtaining creatinine is from creatine, by exposing a mixture of the latter and of hydrochloric acid to evaporation, till all excess of acid is volatilized. The base is best separated from the hydrochlorate, which is thus formed, by digestion with hydrated oxide of lead. The mode of preparing creatinine from urine has been already indicated in our remarks on creatine; moreover, when it is to be prepared from the juice of flesh, the chloride of zinc compound must be employed and decomposed by hydrated oxide of lead; the creatinine may then be readily separated from the creatine by alcohol.

*Tests.*—This body may generally be distinguished with facility from other animal substances, when it is separated as much as possible from

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 257–290.

adherent organic substances. Its alkaline reaction, its property of forming crystalline compounds with the above-named metallic salts, the easy solubility of the compounds which it forms with bichloride of platinum and similar salts, are more than sufficient to characterize it.

*Physiological Relations.*

*Occurrence.*—It is only in the *muscles* and in the *urine* that Liebig has found creatinine. Regarding the quantity in which it exists, nothing is yet known, except that from Liebig's investigations it appears that in the muscles there is far more creatine than creatinine, while in the urine the amount of creatinine very much exceeds that of creatine.

According to Scherer<sup>1</sup> it is highly probable that the *Liquor Amnii* contains creatinine.

*Origin.*—From the facts which have already been communicated it can hardly be doubted that creatinine is produced from creatine; for even if Liebig had not afforded the most decisive proof, by the artificial conversion of one substance into the other, the facts that they occur in an inverse ratio in muscle and in urine, and that putrid urine yields no creatine, but only creatinine, tend to show that also, in the living body, the latter substance proceeds from the former, and consequently is to be regarded purely as a product of excretion.



*Properties.*—This body forms silky, glistening, dazzlingly white needles, is of very difficult solubility in water, and is altogether insoluble in alcohol and ether; it dissolves readily in alkaline solutions, and enters into combination with acids, with the exception of acetic acid.

*Composition.*—This body was discovered and first analyzed by Liebig:<sup>2</sup> its constitution has been recently more accurately determined by Hinterberger<sup>3</sup> under Liebig's superintendence; it consists of:

Carbon, . . . . .	18 atoms,	. . . . .	59.67
Hydrogen, . . . . .	11 “	. . . . .	6.08
Nitrogen, . . . . .	1 “	. . . . .	7.78
Oxygen, . . . . .	6 “	. . . . .	26.52
			100.00

If tyrosine be treated with boiling nitric acid it yields, according to Strecker<sup>4</sup> a large quantity of oxalic acid; but when treated with cold nitric acid it not only yields oxalic acid, but also nitrate of nitro-tyrosine  $\text{C}_{18}\text{H}_{11}\text{N}_3\text{O}_{16}$ ; which when treated with oxide of silver and ammonia yields  $3\text{AgO} + 3\text{HO} + \text{C}_{30}\text{H}_{17}\text{N}_4\text{O}_{17}$ .

*Preparation.*—Cheese, well pressed and freed from adherent butter, or well-dried fibrin or albumen, must be fused, according to Liebig and Bopp,<sup>5</sup> with an equal weight of hydrated potash, till, in addition to ammonia, hydrogen begins to be developed, or, in other words, till the

<sup>1</sup> Zeitschr. f. wissenschaftl. Zoologie. Bd. 1, S. 91.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 127.

<sup>4</sup> Ibid. vol. 72, p. 70-80.

<sup>3</sup> Ibid. Bd. 71, S. 70-79.

<sup>5</sup> Ibid. Bd. 69, S. 19-37.

original dark-brown color merges into a yellow; on then dissolving the mass in hot water, and slightly supersaturating it with acetic acid, the tyrosine separates in needles, which are obtained in a state of perfect purity by solution in potash-water and a second acidulation with acetic acid. The adherent brownish-red pigment may be removed by treating the hydrochlorate of tyrosine with animal charcoal, and boiling the colorless fluid with an excess of acetate of potash; chloride of potassium is then formed, and the tyrosine, free from acetic acid, separates on cooling, in finely matted needles. This substance is also formed, together with leucine and several acids of the first group, during the putrefaction of albumen, fibrin, and casein. Since tyrosine is also formed in the decomposition of the above-named protein-compounds by concentrated hydrochloric acid—or by sulphuric acid (in which latter case leucine is also formed), this mode of procedure may also be adopted for the preparation of this substance. For this purpose we dissolve 1 part of the protein-compound in 4 times the quantity of concentrated hydrochloric acid, and then add 4 parts of sulphuric acid, and evaporate in the water-bath. The hydrochloric acid is expelled by evaporation, from the syrupy, blackish-brown residue, which is then dissolved in water and boiled with milk of lime; the excess of lime is removed from the filtered fluid by sulphuric acid, whose excess is removed by acetate of lead, and the lead by sulphuretted hydrogen: in this syrup crystals of tyrosine and leucine are formed, which are separated from one another in the manner already described.

[In addition to the sources of tyrosine mentioned above (namely casein, albumen, and fibrin), it may be obtained from horn (Hinterberger), cochineal (Warren de la Rue),<sup>1</sup> and from feathers, hair, the elytra of the cockchafer, globulin, and hæmatin (Leyer and Köller),<sup>2</sup> by treatment with dilute sulphuric or concentrated hydrochloric acid, as well as by putrefaction. According to Hinterberger, tyrosine is much more advantageously obtained from cows' horn than from albumen, casein, &c., and it is better to fuse the horn with caustic potash than to employ dilute sulphuric acid. According to Piria,<sup>3</sup> the following is the best method of obtaining this body: 500 grammes of horn shavings must be gradually added to a mixture of 3 litres [5·3 pints] of water and 1300 grammes of sulphuric acid, which must be previously raised to the boiling-point; and the whole must be kept boiling for forty-eight hours; after dilution with much water and neutralization with milk of lime, we treat the filtrate with a little more milk of lime and allow it to boil for an hour or two in order to decolorize it; the fluid, after filtration, is then evaporated to 2½ litres, a stream of carbonic acid being passed continuously through it during the process; on being again filtered and allowed to stand the tyrosine separates in crystals. Leyer and Köller employ the following method: they boil 1 part of protein-substance with 4 parts of sulphuric acid and 12 parts of water for forty hours; the brown fluid is rendered alkaline by milk of lime, and is again heated and filtered. Sufficient sulphuric acid is then added to nearly destroy the alkaline reaction; the tyrosine now crystallizes in tolerable purity from the evaporated filtrate.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 127.

<sup>2</sup> Ibid. vol. 83, p. 332–338.

<sup>3</sup> Ibid. vol. 82, p. 251.

With regard to *testing* for tyrosine, when its quantity is not sufficient to enable us to recognize its presence from its properties, and by its analysis, Piria recommends the employment of the reaction of the salts of tyrosine-sulphuric acid with neutral perchloride of iron, when a dark violet color is manifested. If we place a little tyrosine (a few milligrammes are sufficient) in a watch-glass, moisten it with 1 or 2 drops of sulphuric acid, dilute it after half an hour with water, saturate it when heated with carbonate of lime, and add perchloride of iron (without any free acid) to the filtered fluid, the presence of tyrosine is indicated by the appearance of a dark violet color.—G. E. D.]

### LEUCINE.— $C_{12}H_{13}NO_4$ .

*Properties.*—It occurs in the form of glistening, colorless leaves, which craunch between the teeth, and convey to them the sensation of a fatty matter; it is devoid of taste or odor, is lighter than water, fuses at above  $100^\circ$ , sublimes unchanged when carefully heated to  $170^\circ$ , is soluble in 27.7 parts of water at  $17^\circ.5$ , and in 625 parts of alcohol of 0.828 specific gravity, and in much smaller quantities of hot water and alcohol, but is insoluble in ether; it has no reaction on vegetable colors. No reagent, with the exception of nitrate of suboxide of mercury, precipitates it from its aqueous solution. It dissolves more readily in a solution of caustic ammonia than in water. It dissolves unchanged in concentrated sulphuric and hydrochloric acids, and the solution may even be warmed without the occurrence of decomposition; it dissolves unchanged in cold nitric acid, but, on boiling, is entirely converted into volatile products.

One hundred parts absorb about 28 parts of hydrochloric acid gas. Chlorine gas destroys it. On heating its aqueous solution with nitric oxide or any other oxidizing agent, *leucic acid*,  $C_{12}H_{11}O_5.HO$ , is formed, nitrogen being developed.

If, on the other hand, it is fused with hydrated potash, there is a simultaneous formation of carbonic acid, hydrogen, and valerianate of ammonia ( $C_{12}H_{13}NO_4 + 3KO + 3HO = 2KO.CO_2 + H_3N + 4H + KO.C_{10}H_9O_3$ ). It undergoes the same decomposition during the putrefaction which a solution of pure leucine very readily undergoes when a small quantity of muscular fibre or of albumen has been added.

*Composition.*—Mulder, following Braconnot's investigations regarding leucine, has recently analyzed it, and from his analyses has deduced the formula  $C_{12}H_{12}NO_4$ ; but still later analyses, instituted almost simultaneously by Laurent and Gerhardt,<sup>1</sup> by Cahours,<sup>2</sup> and by Horsford, indicate that in leucine there is contained 1 equivalent of hydrogen more than Mulder had assumed, and continues to assume, in his most recent investigations.<sup>3</sup> Hence leucine, which, moreover, crystallizes without water of crystallization, contains:

<sup>1</sup> Compt. rend. T. 27, pp. 256–258.

<sup>2</sup> Scheikund. Onderzoek. D. 5, pp. 371–377.

<sup>3</sup> Ibid. pp. 265–278.



Carbon,	.	.	.	.	.	12 atoms,	.	.	.	54.96
Hydrogen,	.	.	.	.	.	13	"	.	.	9.92
Nitrogen,	.	.	.	.	.	1	"	.	.	10.68
Oxygen,	.	.	.	.	.	4	"	.	.	24.44
										<hr/>
										100.00

Its atomic weight = 1637.5.

Since leucine possesses scarcely any basic properties, the view that it is a conjugated ammonia =  $\text{H}_2\text{N} \cdot \text{C}_{12}\text{H}_{10}\text{O}_4$ , is the least probable hypothesis regarding its theoretical composition. From Liebig's<sup>1</sup> experiment, to which we have already alluded, that leucine with hydrated potash yields valerianic acid besides volatile products, no theoretical formula for this body can be provisionally deduced; but Gerhardt and Laurent, as well as Cahours, have in part proved it to belong to the series of homologous bodies with the formula  $\text{C}_n\text{H}_{n+1}\text{NO}_4$ , to which, as we shall presently see, sarcosine and glycine pertain.<sup>2</sup> But Cahours,<sup>2</sup> and subsequently Strecker,<sup>3</sup> availed themselves of Piria's mode of proceeding, by which he decomposed the amide-compounds by nitric oxide (see p. 44) into water, nitrogen, and the original acid, in order to obtain the above-mentioned *leucic acid* from leucine. According to this view, leucine should be regarded as *the amide* of this acid: since  $\text{H}_4\text{NO} \cdot \text{C}_{12}\text{H}_{11}\text{O}_5 - 2\text{HO} = \text{C}_{12}\text{H}_{13}\text{NO}_4$ , the theoretical formula for this substance must be  $\text{H}_2\text{N} \cdot \text{C}_{12}\text{H}_{11}\text{O}_4$ .

*Combinations.*—According to Gerhardt and Laurent, leucine, in combination with acids, yields very beautifully crystallizable salts, but they bear much more the character of conjugated acids, so that we might regard leucine in itself as an adjunct; against which view, however, it may be observed that here the adjunct loses no water, as in other cases it usually does on entering into combination, and on separation takes up no water; these combinations are, however, not to be compared with the acid oxide-of-ethyl salts, since only one atom of acid ever combines with leucine; they are, in one respect, most similar to those others which may be equally represented as true neutral salts or conjugated acids, as, for instance, the salicylates of oxide of methyl and of oxide of ethyl; but still more to the compounds of the alkaloids with neutral metallic salts, such as we treated of in our remarks on creatinine.

*Nitrate of leucine, leuconitric acid*,  $\text{C}_{12}\text{H}_{13}\text{NO}_4 \cdot \text{HO} \cdot \text{NO}_5$ , separates in crystals on saturating moderately concentrated nitric acid with leucine; it has an acid but not sharp taste; the salts decrepitate on being heated, and some of them are crystallizable.

*Hydrochlorate of leucine*,  $\text{C}_{12}\text{H}_{13}\text{NO}_4 \cdot \text{HCl}$ , also crystallizes readily.

*Leucic acid*,  $\text{C}_{12}\text{H}_{11}\text{O}_5 \cdot \text{HO}$ , is not only formed in the above manner by oxidizing agents on leucine, but also, when an aqueous solution of this substance has been for a long time exposed to the air, it then develops a nauseous odor, and in the solution we find the ammonia-salt of this acid. It is not crystalline, but oleaginous, dissolves freely in alcohol and ether, and forms crystallizable salts with bases.

Cahours has pointed out the analogy of leucine with the base *thialdine*, discovered by Liebig and Wöhler;<sup>1</sup> both bodies containing the same

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 128.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 52-55.

<sup>3</sup> Compt. rend. T. 27, pp. 265-268.

<sup>4</sup> Ibid. Bd. 61, S. 1-11.

equivalents of carbon, hydrogen, and nitrogen, and the 2 atoms of oxygen of the leucine being replaced by 2 atoms of sulphur in thialdine. This body is produced when aldehyde-ammonia is brought into contact with caustic ammonia and sulphuretted hydrogen; it forms large, colorless, rhombic tablets, which fuse readily, but again solidify at  $42^{\circ}$ , volatilize when exposed to the air, and can be distilled unchanged in the presence of water, but not in the dry state; they are slightly soluble in water, but dissolve readily in alcohol, and still more so in ether, and exhibit no reaction on vegetable colors. The salts that have been examined are  $C_{12}H_{13}NS_4.HCl$  and  $C_{12}H_{13}NS_4.HO.NO_5$ ; this substance also forms compounds perfectly analogous to those of leucine. On dry distillation with hydrated potash its behavior is very different from that of leucine, since it yields leucoline (otherwise called chinoline).

*Preparation.*—According to Mulder, the *caseous oxide* discovered by Proust, and Braconnot's *aposepidine*, are perfectly identical with leucine. It is principally formed in the putrefaction of casein (Iljenko<sup>1</sup> and Bopp),<sup>2</sup> and of gluten (Walter Crum.)<sup>3</sup> If casein, or any other albuminous body, be fused with equal parts of hydrated potash, and the tyrosine extracted from the dissolved mass in the manner already described, the leucine crystallizes from the mother-liquid, and is readily purified by recrystallization from alcohol. If gelatin be treated in a similar manner, or boiled for a long time in potash lye, we obtain leucine and glycine after saturating with sulphuric acid and removing the sulphate of potash by alcohol; and as glycine is far the less soluble of the two in alcohol, the substances may be thus easily separated from one another. Leucine is, however, also formed by the action of concentrated sulphuric or hydrochloric acid on albuminous substances; if, for instance, flesh be gently warmed with an equal volume of concentrated sulphuric acid, then boiled for nine hours with double its weight of water, the acid saturated with lime, and the residue of the filtered solution extracted with alcohol, we obtain on evaporation impure crystals of leucine, which must be purified by recrystallization. On fusing equal parts of a protein-compound and hydrated potash, but interrupting the operation before the mass has become yellow (as was necessary for the preparation of tyrosine), we obtain only leucine according to the method given for tyrosine, since the latter seems to be formed from the former by prolonged action.

*Tests.*—If the leucine be obtained in a state of tolerable purity, and the properties coincide with those known to pertain to leucine, its decomposition into valerianic acid, &c., and its behavior with nitric acid, afford tolerably certain means of distinguishing it. An elementary analysis might, however, be not altogether superfluous, since it may be expected that there are a number of similar bodies for whose discovery and detailed description we may daily look.

#### SARCOSINE.— $C_6H_7NO_4$ .

*Properties.*—Broad, colorless, transparent plates or right rhombic prisms, acuminate on the ends by surfaces set perpendicular on the

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 264–273.

<sup>2</sup> Ibid. Bd. 69, S. 19–37.

<sup>3</sup> Berzelius, Lehrb. d. Ch. Bd. 9, S. 684.

obtuse angles, melting at  $100^{\circ}$ , and subliming unchanged at a somewhat higher temperature. Sarcosine is extremely soluble in water, sparingly soluble in alcohol, and insoluble in ether; the aqueous solution has a sweetish, sharp, faintly metallic taste, has no action on vegetable colors, and is not affected by nitrate of silver or corrosive sublimate; with salts of the oxide of copper it yields the same deep blue color as is produced by ammonia. According to Laurent and Gerhardt,<sup>1</sup> when fused with hydrated potash, it yields, like leucine, hydrogen, ammonia, and carbonic acid, but acetic in place of valerianic acid. ( $\text{C}_6\text{H}_7\text{NO}_4 + 3\text{KO} + 3\text{HO} = 2\text{KO}.\text{CO}_2 + 4\text{H} + \text{H}_3\text{N} + \text{KO}.\text{C}_4\text{H}_3\text{O}_3$ .)

*Composition.*—For the discovery and analysis of this body we are also indebted to Liebig. In accordance with the above formula calculated by Liebig,<sup>2</sup> it consists of:

Carbon,	.	.	.	.	6 atoms,	:	.	.	.	40.45
Hydrogen,	.	.	.	.	7 "	.	.	.	.	7.86
Nitrogen,	.	.	.	.	1 "	.	.	.	.	15.73
Oxygen,	.	.	.	.	3 "	.	.	.	.	35.96
										<hr/> 100.00

Its atomic weight = 111.25.

It is worthy of remark that this body is isomeric with the *lactamide* discovered by Pelouze (see p. 88), and the *urethane* prepared by Dumas from chloro-carbonic ether; hence it is the more important to ascertain the theoretical composition or the proximate grouping of the atoms in these bodies. We might take the commonly accepted view that lactamide is amide with lactic acid deprived of one atom of oxygen =  $\text{H}_2\text{N}.\text{C}_6\text{H}_5\text{O}_4$ , and according to the hypothesis of Berzelius, regard sarcosine as a conjugated ammonia =  $\text{H}_3\text{N}.\text{C}_6\text{H}_4\text{O}_4$ , which indeed is the most probable; but it is worthy of remark that lactamide, as has already been observed in p. 88, is exhibited from lactide (a body isomeric with the adjunct of ammonia in sarcosine) and ammonia; hence we should have anticipated the formation of sarcosine, but not that of an amide. The disintegration of lactamide by potash into lactic acid and ammonia, and on the other hand that of sarcosine into acetic acid, &c., would in itself be sufficient to show that these bodies were differently constituted, even if their other properties did not prove it. If, as Laurent and Gerhardt, as also Cahours,<sup>3</sup> expect, sarcosine is actually decomposed by nitric oxide into lactic acid, then, seeing that we are acquainted with actual lactamide, Piria's test for amide would not prove very much, and the evidence of the amide nature of leucine and of glycine (which we are about to describe) would fall to the ground.

*Combinations.*—Sarcosine forms very crystallizable salts with several acids.

*Hydrochlorate of sarcosine*,  $\text{C}_6\text{H}_7\text{NO}_4.\text{HCl}$ , crystallizes in small, transparent needles and granules; its solution, like that of the hydrochlorate of creatinine, yields no precipitate with bichloride of platinum, but on evaporation we obtain a soluble double compound,  $\text{C}_6\text{H}_7\text{NO}_4$ .

<sup>1</sup> Compt. rend. T. 27, pp. 256-258.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 272.

<sup>3</sup> Compt. rend. T. 27, pp. 265-268.

$\text{HCl} + \text{PtCl}_2 + 2\text{H}_2\text{O}$ , which crystallizes in honey-colored octohedral segments.

*Sulphate of Sarcosine*,  $\text{C}_6\text{H}_7\text{NO}_4 \cdot \text{HO} \cdot \text{SO}_3 + \text{Aq.}$ , crystallizes either in large, feathery plates, or in four-sided, strongly lustrous prisms; it is soluble in water and hot alcohol, and reddens litmus.

With *acetate of copper* sarcosine yields a deep, dark blue, double salt, which crystallizes in thin plates.

*Preparation.*—This base has not yet been found preformed in the animal body, and is only known as a product of the decomposition of creatine, from which it is obtained in the following manner. If a boiling saturated solution of creatine be digested with pure crystallized caustic baryta, in the proportion of ten parts by weight of baryta to one part of creatine, and, after ammonia ceases to be developed, the carbonate of baryta is removed by filtration, sarcosine will separate in crystals from the filtrate; it must be purified by the precipitation of its sulphate by alcohol, and by the decomposition of this salt by carbonate of baryta.

*Tests.*—The mode in which it is obtained and the properties which we have described, afford sufficient evidence to identify their substance.

#### GLYCINE.— $\text{C}_2\text{H}_5\text{NO}_2$ .

*Properties.*—This body, which was formerly named *sugar of gelatin*, and has more recently been known as *glycocoll*, crystallizes in colorless rhombic prisms belonging to the monoclinometric system, which craunch between the teeth, taste less sweet than cane-sugar, and are devoid of odor; these prisms are unaffected by exposure to the atmosphere; at  $100^\circ$  they lose no water; at  $178^\circ$  they melt and become decomposed; they dissolve in 4.3 parts of cold water, more difficultly in cold, but more easily in hot spirit of wine; they are almost insoluble in absolute alcohol and quite so in ether; these solutions have no effect on a ray of polarized light or on vegetable colors. Exposed to the action of the galvanic circuit glycine is very readily decomposed, at the negative pole there being an alkaline reaction from the separation of ammonia, while at the positive pole there is an acid reaction. Glycine dissolves unchanged in the mineral acids, and in alkaline solutions, if not too concentrated. Sulphate of copper and potash yield with glycine a deep blue solution from which no suboxide of copper separates on the application of heat. Further, on boiling glycine with a concentrated solution of potash, or with hydrated baryta or oxide of lead, the fluid developes ammonia and assumes a brilliant fiery red tint, which, however, disappears on the prolonged application of heat. In this process, in addition to the ammonia, there are formed, hydrogen, oxalic acid, and hydrocyanic acid (Horsford). If on the other hand it be fused with hydrated potash, it undergoes a decomposition analogous to that of leucine and sarcosine, into formic acid, ammonia, carbonic acid, and hydrogen gas ( $\text{C}_2\text{H}_5\text{NO}_2 + 3\text{KO} \cdot \text{H}_2\text{O} = 2\text{KO} \cdot \text{CO}_2 + 4\text{H} + \text{KO} \cdot \text{C}_2\text{HO}_3$ . Gerhardt and Laurent).<sup>1</sup> If, finally, an aqueous solution of glycine be treated with nitrous acid or nitric oxide, glycolic acid =  $\text{C}_2\text{H}_3\text{O}_5 \cdot \text{HO}$  (Strecker),<sup>2</sup> is formed, nitrogen gas

<sup>1</sup> Compt. rend. T. 27, pp. 256-258.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 54.

being developed. Moreover, a non-nitrogenous acid, which in all probability is identical with glycolic acid, is produced by chlorine gas and other strongly oxidizing influences, as, for instance, hypermanganate, nitrate, and chlorate of potash (Horsford).

Horsford has analyzed the baryta-salt, and deduced for the acid the formula  $C_3H_3O_6$ , but the analysis yielded less hydrogen and more carbon than are represented by this formula; if Horsford had accidentally omitted to calculate for the organic substance the carbonic acid retained in the baryta, the formula of the baryta-salt would be  $BaO.C_4H_3O_6$ , and consequently would correspond with that of Strecker's acid. The baryta-salt was somewhat insoluble, but crystallized well.

*Composition.*—According to the above formula which is deduced from the coincident analyses of Laurent,<sup>1</sup> Mulder,<sup>2</sup> and Horsford, free glycine, dried at  $100^\circ$ , consists of:

Carbon,	.	.	.	.	4 atoms,	.	.	.	.	32.00
Hydrogen,	.	.	.	.	5 "	.	.	.	.	6.67
Nitrogen,	.	.	.	.	1 "	.	.	.	.	18.67
Oxygen,	.	.	.	.	3 "	.	.	.	.	42.66
										<hr/> 100.00

Its atomic weight = 937.5. Horsford,<sup>3</sup> who has recently made the most complete investigation regarding this substance, is led, from a consideration of its compounds with acids, as well as with certain metallic oxides, to assign to free glycine the formula  $C_4H_4NO_3.HO$ , regarding it as containing 1 atom of combined water; thus throwing doubts upon the homology of leucine, sarcosine, and glycine, maintained by Laurent and Cahours. The analogy in the constitution of these three bodies is undeniable; independently of the fact that the empirical formula  $C_nH_{n+1}NO_4$  is also applicable to hydrated glycine, its relation towards hydrated potash as well as towards nitric oxide, indicates its extreme similarity to the two other bodies. Strecker's discovery that glycolic acid is produced when glycine is decomposed by nitric oxide would lead to the inference that glycine is the amide of glycolic acid, just as leucine might be regarded as the amide of leucic acid. Berzelius\* assumes for glycine double the above atomic weight, and hence he writes its empirical formula  $= C_8H_8N_2O_6 + 2HO$ ; theoretically he regards it as an alkaloid, namely, as ammonia conjugated with a nitrogenous body, so that its rational formula is  $H_3N.C_8H_8NO_6 + 2HO$ .

Here, indeed, the homology with sarcosine entirely fails. Berzelius bases his view regarding the establishment of the doubled atomic weight on the strong acidity of the salts containing 1 atom of this acid,  $C_4H_4NO_3$ ; but in such weak basic bodies, little stress should be laid on this acidity, while, moreover, the compound of glycine with salts, and especially with chlorides, entirely supports the atomic weight assigned by Horsford. It is chiefly from the behavior of glycine when acted on by the galvanic current that Horsford is inclined to regard it as a salt-like compound, namely, as a compound isomeric with the hypothetical anhydrous fumarate of ammonia, since  $C_4H_4NO_3 = H_3N + C_4HO_3$ . Probably, however, Laurent and Strecker's hypothesis still holds good, since, in organic

<sup>1</sup> Compt. rend. T. 20, p. 789.

<sup>2</sup> Journ. f. pr. Ch. Bd. 28, S. 294-297.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 60, S. 1-57.

<sup>4</sup> Jahresber. Bd. 27, S. 655.

nature, we much more frequently meet with amide-compounds than with compounds of anhydrous acids with ammonia.

*Combinations.*—All the combinations of glycine with acids are crystallizable, of tolerable easy solubility, and have a strong acid reaction.

*Neutral hydrochlorate of glycine*,  $C_4H_4NO_3.HO.HCl$ , crystallizes in long flat prisms, which are transparent and glistening, soon deliquesce when exposed to the atmosphere, and dissolve readily in water and in spirit of wine, but slightly in absolute alcohol. Horsford has prepared the following basic hydrochlorates:— $2C_4H_4NO_3 + HO + HCl$ , rhombic prisms not affected by the atmosphere;  $2(C_4H_4NO_3.HO) + HCl$ , which crystallizes well;  $3C_4H_4NO_3 + 2HO + 2HCl$  was obtained from dry glycine in hydrochloric acid gas; in a similar way the same salt was obtained with only 1 atom of water: these basic salts might possibly be mixtures of two salts. Berzelius<sup>1</sup> obtained a combination of hydrochlorate of glycine and *bichloride of platinum*, by extracting a mixture of these two compounds with absolute alcohol, and then precipitating the excess of hydrochlorate of glycine from the solution by ether; the double compound which he thus obtained, occurred in the form of yellow, oily drops, which when exposed to the air crystallized in yellow needles like wavellite; this compound is easily soluble in water and in alcohol, and contains much water of crystallization, in which respects it is very different from the analogous double compounds of most of the alkaloids. If, however, free glycine be mixed with bichloride of platinum, a compound is formed which is represented by  $C_4H_4NO_3 + 2HO + PtCl_2$ , and occurs in black (Berzelius) or red crystals (Horsford). The following compounds with sulphuric acid were obtained by Horsford:  $C_4H_4NO_3.SO_3$ ;  $C_4H_4NO_3.HO.SO_3$ ;  $3(C_4H_4NO_3.HO) + 2(SO_3.HO)$ ;  $3C_4H_4NO_3 + 2SO_3 + HO$ ;  $3(C_4H_4NO_3.HO) + 2SO_3 + HO$ .

*Nitrate of glycine*,  $C_4H_4NO_3.HO + NO_5.HO$ , usually occurs in the form of acicular crystals, but sometimes as large tabular crystals of the monoclinometric system; these crystals are unaffected by exposure to the atmosphere and have an acid taste.

Nitrate of glycine was formerly regarded as a conjugated acid, but these compounds which result from the union of nitrate of glycine with bases, are true nitrates, since, as Horsford has shown, they are directly produced on digesting the nitrates with glycine.

*Oxalate of glycine*,  $C_4H_4NO_3.HO.C_2O_3$ , occurs in wavellite-like crystals, which are unaffected by exposure to the atmosphere.

*Acetate of glycine*,  $C_4H_4NO_3.HO.C_4H_3O_3 + 2HO$ , is crystallizable, and insoluble in alcohol.

Horsford further observed that glycine formed crystallizable compounds with many salts (similar to that which it forms with bichloride of platinum), most of which contain 1 atom of glycine to 1 atom of the salt. With *bases*, especially with hydrated baryta and potash, crystallizable compounds are also formed. *Protoxide of copper-glycine* was obtained by Boussingault, and found to be represented by the formula  $C_4H_4NO_3.CuO$ ; Horsford found 1 atom of water in this compound which crystallized in brilliant blue needles. Similarly to the hydrated oxide of copper, the hydrated oxide of lead, and oxide of silver, may be dis-

<sup>1</sup> Jahresber. Bd. 27, S. 658.

solved in an aqueous solution of pure glycine, and the compound, after being precipitated by the addition of alcohol, may be obtained in a crystalline form. The *lead-compound* crystallizes in prisms, the *silver-compound* in wart-like masses.

There is much regarding these compounds that still remains to be investigated; we have, however, entered more fully into the subject of their composition than we should otherwise have done, because it is on this point that we must form our judgment respecting the constitution of glycine, and decide in favor of one or the other of the above hypotheses.

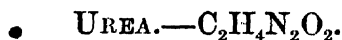
*Preparation.*—Glycine has not yet been found in an isolated state in the animal body: there is, however, reason for believing that this substance is contained preformed as an adjunct in certain known animal acids; moreover, the relations of this body towards acids, bases, and salts (which we have already described), support this view; while, in many cases with which we shall become acquainted as we proceed, it is more than probable that glycine is formed on the separation of the acid from its proper adjunct, as glycerine is produced in the saponification of the hypothetical oxide of lipyl. As instances, we may mention hippuric and glycocholic acids; and when we treat of these acids, we shall enter into the physiological relations and the genesis of glycine.

It has long been known that glycine is a product of the decomposition of animal substances, especially of gelatin, by the action of concentrated mineral acids or caustic alkalis. The following is the best method of obtaining it from gelatin. If the gelatin be boiled with a strong solution of potash till ammonia ceases to be developed, it becomes entirely decomposed into a mixture of 4 parts of glycine and 1 of leucine; the fluid neutralized with sulphuric acid is evaporated to dryness, and the residue extracted with spirit of wine which dissolves both the glycine and the leucine; the glycine, as being the least soluble in alcohol, crystallizes first, while the leucine subsequently crystallizes; by recrystallization and treatment with a little animal charcoal, the glycine can be obtained perfectly pure.

The method of obtaining glycine from hippuric acid is even simpler; for if 1 part of this acid be boiled for half an hour with 4 parts of concentrated hydrochloric acid, it becomes decomposed into glycine and benzoic acid; on the addition of water to the boiled fluid, a great part of the benzoic acid separates and must be removed by filtration; the clear fluid is then evaporated nearly to dryness, and the residue (hydrochlorate of glycine) decomposed with caustic ammonia; finally the glycine is precipitated by, and washed with, absolute alcohol.

*Tests.*—When the substance suspected to be glycine is separated as much as possible from all other matters, the most striking of the properties by which it may be distinguished are its relation towards a hot solution of potash, its difficult solubility in alcohol, and the blue solution which it yields with caustic potash and sulphate of copper, without any separation of the suboxide; and if, further, we study its power of combining with acids as well as with baryta, oxide of copper, oxide of lead, &c., and forming crystallizable bodies, there can hardly remain any doubt regarding its nature. It may easily be distinguished from leucine by the form of its crystals and by its becoming decomposed on exposure to heat.

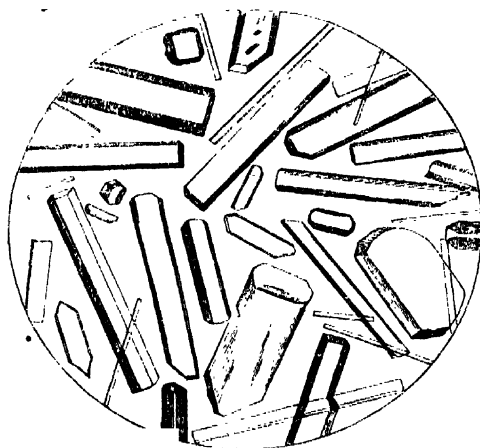
According to Horsford the quantities of urea and uric acid in the urine are increased after the ingestion of glycine, but no unchanged glycine is found in the urine.



*Chemical Relations.*

*Properties.*—Urea crystallizes, when it separates rapidly, in white, silky, glistening needles; but when the crystallization is effected slowly, in flat, colorless, four-sided prisms full of cavities and appearing to be formed of numerous parallel crystalline lamellæ: at the ends the prism

Fig. 6.



Urea, prepared from urine, and crystallized from aqueous solution by slow evaporation.

is terminated by one or two oblique surfaces. According to C. Schmidt,<sup>1</sup> these forms do not pertain to the monoclinometric system, but rather to a hemihedral form belonging to the rhombic system, and bounded by parallel surfaces. These crystals contain no water. Urea is devoid of smell, of a saltish, cooling taste, and is unaffected by exposure to the atmosphere; it dissolves readily in its own weight of water, giving rise to a marked evolution of heat; in hot water it dissolves in every proportion; it is also soluble in 4 or 5 parts of cold and in 2 parts of warm alcohol; it is insoluble in ether, if anhydrous and devoid of alcohol, and in ethereal oil, and exerts no action on vegetable colors. Its concentrated aqueous solution is not changed by boiling or by long keeping, but a dilute solution suffers change.

At about  $120^\circ$  urea fuses without suffering change, but at a little above that temperature it begins to develop ammonia, to become pulpy, and to change into cyanuric acid ( $3\text{C}_2\text{H}_4\text{N}_2\text{O}_2 = 3\text{H}_3\text{N} + \text{C}_6\text{HN}_3\text{O}_4 \cdot 2\text{HO}$ ); when rapidly heated it also yields cyanic acid which is produced from the previously formed cyanuric acid ( $\text{C}_6\text{HN}_3\text{O}_4 \cdot 2\text{HO} = 3\text{C}_2\text{NO} \cdot \text{HO}$ ). On heating urea very slowly, it becomes converted (according to Wöhler

<sup>1</sup> Entwurf u. s. w. S. 41.



and Liebig<sup>1</sup>) into a glistening white body, insoluble in water but soluble in acids and alkalis, carbonic acid and ammonia being evolved during the process. This body =  $C_4H_6N_4O_2$ , for  $3C_2H_4N_2O_2 - (2CO_2 + 2H_3N) = C_4H_6N_4O_2$ . If, on the other hand, urea be kept for some time in a state of fusion at from  $150^\circ$  to  $170^\circ$ , not only are the above-named compounds formed, but also (according to Wiedemann<sup>2</sup>) the *biuret*,  $C_4H_5N_3O_4$ , whose production is explained by the equation,  $2C_2H_4N_2O_2 - H_3N = C_4H_5N_3O_4$ .

If chloride of sodium or hydrochlorate of ammonia be present in a solution of urea, the former will crystallize in octohedra and the latter in cubes; if, however, the crystals be again dissolved in water, and allowed to crystallize anew, they separate in the ordinary manner, namely, the chloride of sodium into cubes, and the hydrochlorate of ammonia into octohedra or feathery forms.

Urea will combine only with certain acids and a few bases; neither the metallic salts, tannic acid, nor any other reagent, can precipitate it from its solutions.

On heating a concentrated solution of urea with nitrate of silver, cyanate of silver separates, while nitrate of ammonia remains in solution ( $C_2H_4N_2O_2 + AgO.NO_3 = AgO.C_2NO + H_3N.HO.NO_3$ ).

By nitrous acid urea is decomposed into nitrogen, water, and carbonic acid ( $C_2H_4N_2O_2 + 2HO + 2NO_3 = 6HIO + 2CO_2 + 4N$ ); by chlorine into nitrogen, carbonic acid, and hydrochloric acid ( $C_2H_4N_2O_2 + 2HO + 6Cl = 6HCl + 2CO_2 + 2N$ ).

On boiling urea either with strong mineral acids or with caustic alkalis, it takes up 2 atoms of water and is decomposed into ammonia and carbonic acid ( $C_2H_4N_2O_2 + 2HO = 2H_3N + 2CO_2$ ).

If organic matters, either putrefying or capable of undergoing putrefaction, be mixed with an aqueous solution of urea, the latter is soon converted into carbonic acid and ammonia.

*Composition.*—According to the above-formula urea consists of:

Carbon,	.	.	.	.	.	2 atoms,	.	.	.	20.000
Hydrogen,	.	.	.	.	.	4 "	.	.	.	6.666
Nitrogen,	.	.	.	.	.	2 "	.	.	.	46.667
Oxygen,	.	.	.	.	.	2 "	.	.	.	26.667
										<hr/> 100.000

Its atomic weight = 750.0. Although there have been many discussions regarding the rational constitution of urea, much still remains to be cleared up. Dumas, after his discovery of oxamide, started the hypothesis, that urea is an amide of carbonic acid, since  $2H_3N + 2CO_2 - 2HO = C_2H_4N_2O_2$ , and the relation of urea towards nitrous acid, and its ready decomposition into carbonic acid and ammonia, seem to support this view. But Berzelius justly points out the analogy, in their combining relations with acids, between the alkaloids and urea, and regards the latter as ammonia conjugated with a nitrogenous body which he names *urenoxide*, so that the rational formula for urea would be =  $H_3N.C_2HNO_2$ . Independently of the analogy between the salts of urea with those of the alkaloids, the following consideration mainly supports this view: cyanate

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 54, S. 371.

<sup>2</sup> Journ. f. pr. Ch. Bd. 43, S. 271-280.

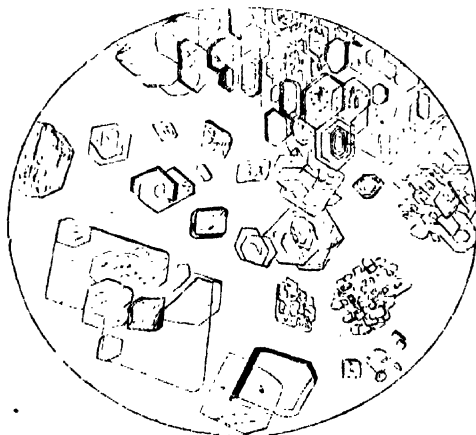
of ammonia =  $\text{H}_3\text{N} \cdot \text{HO} \cdot \text{C}_2\text{NO}$ , is convertible, as we shall presently see, into urea; the grouping of the atoms in urea must be perfectly different from that in this salt, since urea has lost all the properties of a salt. But we know that free hydrated cyanic acid is spontaneously converted by a transposition of its atoms into the so-called *cyamelide* =  $\text{C}_2\text{HNO}_2$ ; now, nothing is more obvious than to assume that in the combination with ammonia the cyanic acid becomes incorporated with the water of the ammonia-salt, in the same manner as in the free state, and that this cyamelide, if not identical with, is highly analogous to the urenoxide of Berzelius, and thus forms the adjunct of the ammonia in urea. Probably, also, the existence of the biuret might be made available in the support of this hypothesis, since the most simple view of the biuret is to regard it as consisting of 2 atoms of urenoxide and 1 atom of ammonia, for  $\text{C}_4\text{H}_5\text{N}_3\text{O}_4 = \text{H}_3\text{N} + 2\text{C}_2\text{HNO}_2$ .

*Combinations.*—It is only with some acids that urea has a tendency to combine. Cap and Henry<sup>1</sup> fancied that they had prepared compounds of urea with sulphuric, lactic, hippuric, and uric acids, but the existence of those compounds is very correctly doubted. We know with certainty only three salts of urea, namely, the hydrochlorate, the nitrate, and the oxalate.

*Hydrochlorate of urea*,  $\text{C}_2\text{H}_4\text{N}_2\text{O}_2 \cdot \text{HCl}$ , was simultaneously obtained by Erdmann<sup>2</sup> and Pelouze.<sup>3</sup> They prepared it by passing a stream of dry hydrochloric acid gas over urea. The compound is white and hard, and crystallizes in plates; it attracts water from the atmosphere, and from this water the hydrochloric acid escapes by evaporation, and pure urea crystallizes; in water the salt becomes rapidly decomposed into hydrochloric acid and urea.

*Nitrate of urea*,  $\text{C}_2\text{H}_4\text{N}_2\text{O}_2 \cdot \text{HO} \cdot \text{NO}_5$  (according to the analysis of Reg-

Fig. 7.



Nitrate of urea, separated from very concentrated human urine by nitric acid.

nault, which has been repeated by Marchand,<sup>4</sup> Heintz,<sup>5</sup> Fehling,<sup>6</sup> and Werther),<sup>7</sup> is formed by mixing a concentrated solution of urea with an

<sup>1</sup> Journal de Pharm. T. 25, p. 133.

<sup>2</sup> Ann. de Ch. et de Phys. 3 Sér. T. 6, p. 63.

<sup>3</sup> Journ. f. pr. Ch. Bd. 35, S. 481.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 55, S. 249.

<sup>5</sup> Journ. f. pr. Ch. Bd. 25, S. 506.

<sup>6</sup> Pogg. Ann. Bd. 66, S. 114-122.

<sup>7</sup> Journ. f. pr. Ch. Bd. 35, S. 51-66.

excess of nitric acid; the compound at once separates (on cooling, almost perfectly) in large nacreous, shining scales, or in small, glistening, white plates; on examining under the microscope the contact of the urea and the nitric acid, we first observe very obtuse rhombic octohedra, at whose acute angles ( $= 82^\circ$ ) more particles are gradually accumulated, so that they appear to increase in size, and the octohedra become converted into rhombic tablets, or form hexagonal tablets (whose opposite acute angles likewise are  $82^\circ$ ); these crystals always occur isolated, or in uniformly superimposed masses (C. Schmidt).<sup>1</sup> This salt is uninfluenced by the atmosphere, has an acid taste, is more soluble in pure water than in water containing nitric acid, and dissolves in alcohol, producing considerable depression of temperature; on evaporating its aqueous solution, the salt very readily effloresces; it reddens litmus; a concentrated solution is not affected by boiling, but a dilute solution is converted into carbonic acid, carbonate of ammonia, water, and nitrous oxide ( $\text{C}_2\text{H}_4\text{N}_2\text{O}_2 \cdot \text{HO} \cdot \text{NO}_5 = \text{H}_3\text{N} + 2\text{CO}_2 + 2\text{HO} + 2\text{NO}$ ). On heating dried nitrate of urea rapidly, it decrepitates, but on heating it slowly to  $140^\circ$ , it becomes decomposed into carbonic acid, nitrous oxide, urea, and nitrate of ammonia. If the solution of this salt be not too dilute, a solution of oxalic acid precipitates oxalate of urea.

*Oxalate of urea*,  $\text{C}_2\text{H}_4\text{N}_2\text{O}_2 \cdot \text{HO} \cdot \text{C}_2\text{O}_3$  (sometimes, according to Marchand, taking up 2 atoms of water of crystallization) is also obtained by the direct union of the constituent parts, and forms, as far as the unaided eye can perceive, long thin plates or prisms; under the microscope it is usually seen in hexagonal plates, similar to those of nitrate of urea, interspersed occasionally with four-sided prisms with planes of truncation proceeding from the broader sides of the rectangular section (Fig. 8). The form of this oxalate, like that of the nitrate of urea, belongs to the monoclinometric system. This salt has an acid taste, dissolves at  $16^\circ$  in 22.9 parts of water and in 62.5 of alcohol; it is precipitated from its aqueous solution by an excess of oxalic acid. On exposure to heat it is decomposed into carbonate of ammonia and cyanuric acid.

Like glycine, urea also unites with *salts*, which hold it in such firm combination, that not only does no decomposition ensue when their solutions are boiled, but even oxalic and nitric acids fail to separate the urea from some of their compounds (Werther).<sup>2</sup>

On mixing concentrated solutions of urea and nitrate of silver, there are formed thick prisms with a rhombic base which are readily soluble in water and alcohol  $= \text{C}_2\text{H}_4\text{N}_2\text{O}_2 \cdot \text{AgO} \cdot \text{NO}_5$ . On the addition of a solution of soda to the solution of these crystals, a yellow precipitate is obtained  $= 5\text{AgO} + 2\text{C}_2\text{H}_4\text{N}_2\text{O}_2$ . Besides these, Werther has also obtained the following combinations:— $\text{C}_2\text{H}_4\text{N}_2\text{O}_2 + 2\text{AgO} \cdot \text{NO}_5$ ;  $\text{CaO} \cdot \text{NO}_5 + 3\text{C}_2\text{H}_4\text{N}_2\text{O}_2$ ;  $\text{MgO} \cdot \text{NO}_5 + 2\text{C}_2\text{H}_4\text{N}_2\text{O}_2$ ;  $\text{NaO} \cdot \text{NO}_5 + \text{C}_2\text{H}_4\text{N}_2\text{O}_2 + 2\text{HO}$ ;  $\text{NaCl} + \text{C}_2\text{H}_4\text{N}_2\text{O}_2 + 3\text{HO}$ , crystallizing in deliquescent rhombic prisms;  $2\text{HgCl} + \text{C}_2\text{H}_4\text{N}_2\text{O}_2$ , flat prisms glistening like mother-of-pearl. Urea cannot be separated from the solutions of these compounds either by nitric or oxalic acid.

*Products of its metamorphosis.*—*Biuret*,  $\text{C}_4\text{H}_6\text{N}_3\text{O}_4$ , is, as we have

<sup>1</sup> Entwurf u. s. w. S. 42–45.

<sup>2</sup> Journ. f. pr. Ch. Bd. 35, S. 51–60.

already mentioned, the chief product (together with cyanuric acid) which is obtained on heating pure urea or its nitrate to a temperature of  $152^{\circ}$ — $170^{\circ}$ ; the cyanuric acid is precipitated by basic acetate of lead from the aqueous solution of the fused product, and the excess of lead removed by sulphuretted hydrogen; the biuret is then obtained by the evaporation of the solution. It forms small crystals which dissolve readily in water, and still more readily in alcohol; it exerts no action on vegetable colors, does not combine with bases, and dissolves unchanged in concentrated sulphuric and nitric acids; with sulphate of copper and potash it yields a red solution. Its rational formula  $= \text{II}_3\text{N} + 2\text{C}_2\text{HNO}_2$ .

*Preparation.*—Urea not only occurs preformed in the animal body, but can also be artificially prepared. When Wöhler made the beautiful discovery that urea was formed by the union of cyanic acid and ammonia, the physiologists of that day who were still imbued with ideas of vital forces, were astonished that a matter which appeared only capable of formation by organic force, could also be formed by the hand of the chemist from so-called inorganic matters. The astonishment of the physiologists has, however, gradually ceased, not only because they have for the most part shaken off their adherence to irrational vital forces, but also because since that time many other substances have been artificially produced, which are identical with, or at all events most similar to previously known organic matters. We have learned to regard urea as one of the most common products of decomposition, not only of natural organic bodies, but also of artificial substances. It would occupy too much of our present space, were we to enumerate all the cases in which urea occurs as a product of the decomposition of a nitrogenous substance; we will here only mention its formation on the union of cyanogen and water, of fulminate of copper and hydrosulphate of ammonia (Gladstone),<sup>1</sup> in the decomposition of allantoine by nitric acid, of creatine by the alkalies, of alloxan by a boiling solution of acetate of lead, &c.

There are various ways in which urea may be obtained from urine, but it is chiefly effected by nitric or oxalic acid; it is more advisable to use the alcoholic extract of urine than the residue left by its direct evaporation; if nitric acid be used, the nitrate of urea must be exposed to due pressure between tiles and filtering paper, and after it has been dissolved in a little water, must be decomposed with carbonate of lead or of baryta; crystals of nitrate of lead or of baryta soon separate from the filtered fluid, which must be evaporated and extracted with alcohol; this alcoholic solution may contain, in addition to urea, a little nitrate of lead, but it takes up no nitrate of baryta; when baryta has been used, the alcoholic solution must be decolorized with animal charcoal; when the salt of lead has been used, the solution is often perfectly colorless after the precipitation of the metal by sulphuretted hydrogen. The urea separates in a crystalline form, on the evaporation of the alcoholic solution.

In order to prepare urea from cyanate of ammonia, we raise a mixture of 28 parts of ferrocyanide of potassium, from which all the water has

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 66, S. 1–5.

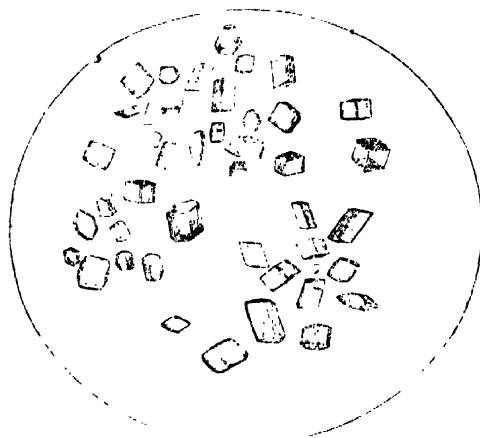
been expelled, and 14 parts of well-dried, good peroxide of manganese, to a faint red heat (even when the mixture is sufficiently heated at a single spot, the whole mass assumes a phosphorescent appearance); from this glowing residue the cyanate of potash which has been formed must be extracted with cold water, and mixed with  $20\frac{1}{2}$  parts of dry sulphate of ammonia; most of the sulphate of potash separates in a crystalline form, while the cyanate of ammonia, now converted into urea, remains in solution. The remaining sulphate is separated by crystallization, but more perfectly by alcohol.

*Tests.*—Urea may generally be very easily recognized by its properties, especially by its behavior towards nitric and oxalic acids; but when we have to discover very minute quantities of this substance in albuminous fluids, it is often very difficult to determine its presence with scientific precision. It is in alcoholic extracts that we must always seek for urea, but before we proceed to search for it, there are several precautionary measures to be adopted, the neglect of which would render our attempt to discover it futile. In the first place, in reference to the presence of albuminous substances, if we wish to discover small quantities of urea in albuminous fluids, we must not be satisfied with the removal of the albumen by simple boiling; since by the coagulation of the albumen the fluid becomes more alkaline, and might, during evaporation, induce a decomposition of the urea; moreover, all albuminous matter is not precipitated by boiling, but a portion remains dissolved by the alkali, and is taken up in the alcoholic extract; on evaporation this albumen undergoes a change which probably co-operates with the alkali in inducing the decomposition of the urea. This may explain how it was that Marchand could only recover 0.2 of a gramme of urea from a mixture of 200 grammes of serum and 1 gramme of urea. Hence, before boiling the albuminous fluid, we must add a few drops of acetic acid, so as to give it a slightly acid reaction, whereby not only is the alkalescence of the fluid prevented, but a much more perfect separation of the coagulable matters is effected. If the residue of the fluid from which the coagulated matters have been filtered be extracted with cold alcohol, and the solution rapidly evaporated, so as to cause the chloride of sodium (taken up by the cold alcohol) to separate as much as possible in crystals, on then bringing a drop of the mother-liquid in contact with nitric acid under the microscope, we shall observe the commencement of the formation of the rhombic octohedra, and the hexagonal tablets, in which, if the investigation is to be unquestionable, the acute angles ( $=82^\circ$ ) must be always measured (Fig. 7). After the determination of the nitrate we may also obtain the oxalate, and submit it to microscopic examination (Fig. 8). A good crystallometric determination yields, however, the same certainty as an elementary analysis which, in these cases, would never, or extremely seldom, be possible.

Formerly the presence of small quantities of urea was supposed to be established when chloride of sodium crystallized in the octohedral form; but independently of the circumstance that other substances besides urea may induce a similar action on the form of the crystals of this salt, it must be borne in mind that chloride of sodium, when we trace the formation of its crystals under the microscope, presents itself in combina-

tions of the regular system, with a complexity varying with the minuteness of the crystals. This occurs when we allow pure chloride of sodium to crystallize: and it is still more the case when organic matters are mixed with the solution. I am acquainted with no other substance of the regular system which presents such uncommon crystals under the microscope as chloride of sodium. We need only expose the alcoholic extract of any animal fluid to spontaneous evaporation, in order to recognize with the naked eye, in the greater crystals, the combinations which we have perceived on examining the crystallization of a solution of pure salt under the microscope.

Fig. 8.



Oxalate of Urea.

In order to determine the *amount* of urea in urine, most analysts have followed the method proposed by Mitscherlich,<sup>1</sup> and have availed themselves of the insolubility of the nitrate. There are several causes of error in this method which cannot be altogether avoided, but with due care may be made very inconsiderable. They chiefly consist in the imperfect insolubility of this salt, and on the adherence of the so-called extractive matters to it; if, however, we use an excess of nitric acid for the purpose of separating the urea, cool the fluid artificially, filter after some time, rinse the salt with cold nitric acid, and, after it has been submitted to pressure, dry it at a temperature not exceeding 110°, we shall not have so great a loss of urea as Heintz<sup>2</sup> maintains must always occur in adopting this method; but in relation to accuracy, the results fall far short of those obtained in the determination of mineral substances. The idea occurred almost simultaneously to Ragsky<sup>3</sup> and Heintz<sup>4</sup> that the urea in urine might be determined quantitatively by its decomposition by *sulphuric acid*. Both investigators have satisfied themselves that the so-called extractive matters of the urine do not modify the result of the experiment; the essential point in this method, which is somewhat more complicated, but doubtless more accurate than that by nitric acid, consists in our determining, by means of bichloride

<sup>1</sup> Pogg. Ann. Bd. 81, S. 303.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 56, S. 29-34.

<sup>2</sup> Ibid. Bd. 66, S. 114-160.

<sup>4</sup> Pogg. Ann. Bd. 68, S. 393-410.

of platinum, the amount of potash and ammonia (if the latter be present) in a specimen of urine, and in our then treating a second specimen with sulphuric acid, and gradually heating it to  $180^{\circ}$  or  $200^{\circ}$ , or as long as any effervescence continues; the fluid is then filtered, and the amount of ammonia determined by bichloride of platinum; deducting from the precipitate thus obtained that which was yielded by the other specimen (corresponding to the potassio-chloride of platinum), we can easily calculate the amount of urea from the ammonio-chloride of platinum, or from the platinum itself left on the incineration of the residue.

A still better method, by which urea may be determined quantitatively, although not perfectly free from error, has been given by Millon.<sup>1</sup> It is based on the fact that urea is decomposed by nitrous acid into nitrogen and carbonic acid; to effect this object a solution of nitrite of suboxide of mercury is dissolved in nitric acid, and added to a weighed portion of urine; on warming this mixture there is a development of nitrogen and carbonic acid, which latter gas is caught in a potash-apparatus and weighed. Some of the extractive matters might yield carbonic acid, even if none of the other constituents of the urine did so; this, however, is denied by Millon. It must also be recollected that the urine always contains free carbonic acid in solution.

Finally, a method has been proposed by R. Bunsen<sup>2</sup> for the quantitative determination of urea, founded on the property that its solutions *undergo decomposition in closed vessels at a temperature of from  $120^{\circ}$  to  $240^{\circ}$* ; the carbonic acid which is thus formed is combined with baryta, and the amount of urea is calculated from that of carbonate of baryta.

### *Physiological Relations.*

*Occurrence.*—Urea is one of the principal products of excretion of the kidneys: hence it chiefly occurs in the urine. Although it constitutes the greatest part of the solid constituents of the urine, it is contained in the liquid urine in very variable quantities in consequence of the physiological relations, in accordance with which the amount of water in the urinary secretion varies in so extraordinary a degree. In order to convince ourselves of the quantity of urea excreted in the urine, we must examine the urine collected in a definite interval in relation to its proportion of urea. As, in the consideration of "Urine," we shall return to this subject, we will here only remark that the urine of a healthy man contains generally from 2.5 to 3.2% of urea, that the ratio of urea to the other solid constituents is about = 9 : 11 or 7 : 9, and that a healthy man in twenty-four hours excretes from 22 to 36 grammes.

My experiments<sup>3</sup> show that the amount of urea which is excreted is extremely dependent on the *nature of the food* which has been previously taken. On a purely animal diet, or on food very rich in nitrogen, there were often two-fifths more urea excreted than on a mixed diet; while, on a mixed diet, there was almost one-third more than on a purely vegetable diet; while, finally, on a non-nitrogenous diet, the

<sup>1</sup> Compt. rend. T. 26, pp. 119-121.    <sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 375-387.

<sup>3</sup> Journ. f. pr. Ch. Bd. 25, S. 22-29, and Bd. 27, S. 257-274.

amount of urea was less than half the quantity excreted during an ordinary mixed diet.

In my experiments on the influence of various kinds of food on the animal organism, and especially on the urine, I arrived at the above results, which in mean numbers may be expressed as follows: on a well regulated mixed diet I discharged, in 24 hours, 32.5 grammes of urea (I give the mean of 15 observations); on a purely animal diet 53.2 grammes (the mean of 12 observations); on a vegetable diet 22.5 grammes (the mean of 12 observations); and on a non-nitrogenous diet 15.4 grammes (the mean of 3 observations).

It is especially worthy of remark, that the augmentation of the urea in the urine occurs very soon after the use of highly nitrogenous food, and that in such cases often five-sixths of the nitrogen taken in the food in 24 hours are eliminated as urea by the kidneys.

When I took 32 boiled hens' eggs daily, I consumed in them about 30.16 grammes of nitrogen, but in the above-mentioned quantity of urea I discharged only about 25 grammes in 24 hours. On the morning following the day on which I had taken only flesh or eggs, the urine was so rich in urea that immediately on the addition of nitric acid it yielded a copious precipitate of nitrate of urea; hence Prout's assertion, may be correct in reference to England, that freshly passed urine often gives a precipitate of nitrate of urea immediately on the addition of nitric acid, although on the continent, where less animal food is taken, no one, so far as I know, has made a similar observation; and hence also the urine of carnivorous animals is very rich in urea (Vauquelin,<sup>1</sup> Hieronymi,<sup>2</sup> Tiedemann and Gmelin),<sup>3</sup> while the urine of gramnivorous animals is comparatively poor in this constituent (Boussingault).<sup>4</sup>

Notwithstanding the considerable influence which the nature of the food exerts on the quantity of urea excreted by the kidneys, there is as much urea in the urine after prolonged absence from all food (after a rigid fast of 24 hours) as after the use of perfectly non-nitrogenous food.

Lassaigne<sup>5</sup> found urea in the urine of a madman who had taken no food for 14 days; and we observe something similar almost daily in patients with typhus fever and other diseases, who for 14 days or more have taken nothing but an oily emulsion or an emollient decoction, and yet always pass urine containing urea, and often rich in it. After living for three days on a perfectly non-nitrogenous diet, I still found, in the morning urine, more than 1% of urea.

Strong *exercise* of the bodily powers causes an increased excretion of urea.

While, from numerous observations, I ascertained that, during my ordinary habits of life, I discharged about 32 grammes in 24 hours, I found that after strong bodily exercise, I, on one occasion, passed 36 grammes, and on another 37.4 grammes in 24 hours.

<sup>1</sup> Schweigg. Journ. Bd. 3, S. 175.

<sup>2</sup> Journ. de Ch. et de Pharm. T. 3, p. 322.

<sup>3</sup> Verdauung u. s. w. Bd. 2, S. 4.

<sup>4</sup> Ann. de Chim. et de Phys. 3 Sér. T. 15, pp. 97-114.

<sup>5</sup> Journ. de Chim. Méd. T. 1, p. 272.



The urine of women and children contains, according to Becquerel,<sup>1</sup> less urea than that of men.

Becquerel found the ratio of urea excreted in 24 hours by women, to that excreted by men = 15·582 : 17·537.

Scherer<sup>2</sup> has obtained much more correct results than Becquerel in the case of children and adults. He found that the urine of young children contained on an average 1·7% of urea, while he found only 1·25% in the 24 hours' urine of a young man aged 22 years. Determinations of this kind lead, however, to few conclusions; to obtain an insight into the general process, our determinations should have reference to definite intervals of time, and to definite weights. A boy aged 3½ years, discharged in 24 hours 12·98 grammes of urea; a girl, aged 7 years, 18·29 grammes; a youth aged 22 years, 37·008 grammes, and a man aged 38 years, 29·824 grammes. If, however, we take the relative weights into consideration, it follows that for 1 killogramme's weight of the child there was discharged 0·810 of a gramme of urea, while for 1 killogramme's weight of the adult, only 0·420 of a gramme (or a little more than half the quantity) of urea was excreted.

Like Becquerel, I have failed in establishing the fact that there is an augmentation of urea in certain forms of disease, although English physicians have shown an inclination to assume a urea-diathesis.

Although we are, *à priori*, prejudiced against all these diatheses which English physicians have attempted to establish on certain urinary analyses, (see p. 54), we must especially protest against such a urea-diathesis; for how does this indicate a morbid process? The nature of this or that disease does not depend on an increased excretion of urea, which is only a consequence of another process. The urea is possibly only excreted in increased quantity when material for its formation is sufficiently supplied; now if polyphagia be not combined with this urea-diathesis, the source of the urea must be sought in the waste or consumption of the nitrogenous tissues; this is not based on the tendency of the tissues to be converted unto urea, but depends on other processes which accompany many morbid processes. In diseases where such a consumption actually occurs, I have never found the urea passed in twenty-four hours exceed the normal quantity, and have very often found it far beneath the average.

A diminution in the amount of urea excreted during disease in twenty-four hours is very frequently observed: this, however, in most cases, may be dependent on the low diet.

Becquerel has made the best observations in reference to this subject; it appears, however, to us, that such investigations may rather serve to enable us to form an opinion of the morbid process in a special case, than to establish general rules regarding the diminution of the urea in the urine in certain classes of disease.

It is by careful observation of the urine in individual cases, and not by drawing general inferences, that we can make these examinations useful.

Many chemists have long sought in vain to detect urea in *normal*

<sup>1</sup> Séméiotique des Urines, &c., Paris, 1841, p. 34.

<sup>2</sup> Verh. d. phys.-med. Ges. z. Würzburg. Bd. 3, S. 180-190.

*blood*; Simon believed that he had found it in calves' blood, and Strahl and Lieberkuhn,<sup>1</sup> and recently Garrod,<sup>2</sup> maintain that they have detected it in human blood: without doubting the correctness of the observations of these chemists, it is only recently that I have been able to convince myself with precision by decisive experiments that urea is present in normal blood.

[Verdeil and Dollfus<sup>3</sup> have found urea in large quantities in the blood of oxen. Moleschott<sup>4</sup> believes that he has found oxalate of urea, together with other oxalates, in the muscular juice of frogs, whose livers had been some days previously extirpated. Grohé<sup>5</sup> has, however, subsequently examined the constituents of the muscular juice of frogs in the Giessen Laboratory, and has arrived at the following results, namely:

1. That neither urea nor oxalic acid exists in this fluid; and
2. That the crystals supposed by Moleschott to consist of oxalate of urea, in reality are composed of creatine, creatinine, and nitrate of potash.—G. E. D.]

In my investigations regarding the amount of alkaline carbonates contained in the blood, I often operated on four or six pounds of fresh ox-blood: in order to avoid the decomposition and re-arrangement of the soluble mineral constituents of blood which always occur in ordinary incineration, I first separated the coagulable matters of the blood, after diluting it with four times its volume of water, and neutralizing it with acetic acid; the residue left by the evaporation of the fluid, from which the coagulated albumen had been removed by filtration, and the films that formed during evaporation had been skimmed off, was treated with absolute alcohol, and then, in the manner we have already described, examined for urea; the measurements of the angles of the crystals both of nitrate and oxalate of urea, which were made according to Schmidt's method under the microscope, exactly coincided with the measurements given by Schmidt for these crystals.

Strahl's method, which I have repeatedly tried, and which consists in the extraction of the urea from four ounces of blood by the addition of alcohol, and in diagnosing the existence of urea from the crystallization of the oxalate, does not appear to me to be sufficiently conclusive; for, in the first place, the quantity of urea in four ounces is very small, even for microscopic observation; secondly, alcohol extracts from the blood certain organic matters which partly separate on evaporation; thirdly, oxalic acid always precipitates mineral matters which render the object indistinct; and, finally, if its crystals be not crystallographically determined, it is often very hard to distinguish oxalate of urea from crystallized alkaline oxalates; all of which reasons led me to think that Strahl's experiments required to be confirmed in some other manner.

Urea increases abnormally in the blood of persons suffering from degeneration of the kidneys, whereby the function of those organs is destroyed. Under the general term of *Bright's disease*, we usually include the various conditions in which there is a mechanical disturbance

<sup>1</sup> Preuss. Vereins-Zeit. No. 47, 1847.

<sup>2</sup> Medico-Chirurgical Transactions, Vol. 31, p. 83.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 74, S. 214.

<sup>4</sup> Arch. f. physiol. Heilk. Bd. 11, S. 493.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 85.

of the urinary secretion, however different the histological alteration in the renal tissue may be; and we use the word *uræmia* to indicate the group of symptoms which depend on the retention of urea in the blood.

Christison<sup>1</sup> was the first who recognized the occurrence of urea in the blood in this disease. In any other disease, urea is only rarely found in the blood; hence, it is by no means requisite that the symptoms of uræmia should be combined with the presence of urea in the blood, since every physician knows how often Bright's disease occurs without this group of symptoms; it is only when the urine is very scanty that these symptoms occur: that of vomiting is not by any means a necessary one, as is generally supposed. Moreover, urea has been found by Rainey<sup>2</sup> and Marchand, in the blood of cholera patients, but only when there was ischuria; and Garrod<sup>3</sup> thinks that he has found it in the blood of a gouty patient.

Rees<sup>4</sup> and Wöhler<sup>5</sup> have detected urea in *Liquor Amnii*, which, they are convinced, contained none of the mother's urine. Mack<sup>6</sup> and Scherer<sup>7</sup> however failed in detecting any urea in this fluid.

[Rees<sup>8</sup> has frequently met with small quantities of urea in milk.—G. E. D.]

Millon<sup>9</sup> found urea in the *vitreous* and *aqueous humors* of the eye, and Wöhler<sup>10</sup> confirms the fact.

Urea has very often been found in *dropsical exudations*.

I have never been able to discover urea in serous exudations, unless at the same time there was disease of the kidneys; previous statements may possibly only have reference to dropsical fluids depending on Bright's disease, and not to those accumulations of fluid which arise from enlargement of the liver.

In Bright's disease, urea is found in all the serous fluids; thus Schlossberger<sup>11</sup> once found it in an aqueous effusion in the cerebral ventricles.

The *matters vomited* in uræmia not unfrequently contain urea. (Nysten<sup>12</sup> and others.)

Wright<sup>13</sup> has found urea in the *saliva* of a patient with Bright's disease, and also in that of a dog poisoned with corrosive sublimate.

Urea has been found by O. B. Kühn in a *biliary concretion*; and Strahl and Lieberkühn have recently detected it in the *bile* after the extirpation of the kidneys.

*Origin*.—Physiologists were long undecided regarding the seat of the actual formation of urea. Since urea had not been discovered in normal blood, many believed that they must adhere to the old view, that the excreta are formed in the excreting organs from the constituents of the blood, and that urea is thus first produced in the kidneys. They

<sup>1</sup> On granular degeneration of the kidneys, &c. Edinburgh, 1839, p. 20.

<sup>2</sup> Lond. Med. Gaz. Vol. 23, p. 518.

<sup>3</sup> Op. cit.

<sup>4</sup> Lond. Med. Gaz. Vol. 23, p. 462.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 98.

<sup>6</sup> Arch. f. phys. u. pathol. Ch. u. Mikr. Bd. 2, S. 218–224.

<sup>7</sup> Zeitschr. f. wissenschaftl. Zoologie. Bd. 1, S. 88–92.

<sup>8</sup> Guy's Hospital Reports, New Series. Vol. 1, p. 328.

<sup>9</sup> Compt. rend. T. 26, p. 121.

<sup>10</sup> Ann. d. Ch. u. Pharm. Bd. 66, S. 128.

<sup>11</sup> Arch. f. phys. Heilk. Bd. 1, S. 43.

<sup>12</sup> Journ. de Chim. Méd. 1837, p. 257.

<sup>13</sup> Lancet, 1844. Vol. 1, p. 150.

accounted for the circumstance that urea is, in certain morbid conditions, sometimes found in the blood and other fluids, by assuming that it was then resorbed from the kidneys or the urinary bladder. To overthrow this opinion, Prevost and Dumas,<sup>1</sup> and subsequently Gmelin, Tiedemann, and Mitscherlich,<sup>2</sup> extirpated the kidneys of animals, and then found no inconsiderable quantity of urea in the blood; indeed Marchand<sup>3</sup> induced all the symptoms of uræmia in a dog by the mere ligature of the renal nerves, and was able to recognize the presence of urea with the greatest certainty, not only in the blood, but also in the vomited matters.

The investigations of Marchand have thrown much light upon this subject; this accurate observer could only recover 0.2 of a gramme of urea from 200 grammes of serum to which 1 gramme of urea had been added; he shows that, even if the urea were only separated from the blood at the end of each successive hour, it could not have accumulated in such quantity as to have been discoverable by the present mode of investigation. The following consideration will give us an idea of the small quantity of urea which, according to Marchand's hypothesis, at the most can accumulate in the blood in one hour. From the experiments of Ed. Weber, which I have in part confirmed, we may assume that there are in an adult man at most 6 or 7 kilogrammes [16 to 19 pounds] of circulating blood; now, if in 24 hours 30 grammes of urea are discharged, at most only 1.25 grammes could accumulate in one hour in the whole mass of the blood, so that only 0.021% could be contained in it; this minute quantity can, however, as we have already shown, only be detected in operating on very large masses of blood, and by the aid of the microscope. Hence it is easy to understand why, during my experiments with an animal diet, while the urine was loaded with urea, none of this substance could be discovered in the blood.

If it be now established, that the urea is not primarily formed in the kidneys, the question still remains to be answered, whether it is produced in the circulating blood or in the individual living organs (as for instance, the muscles), and from what materials it is principally formed. In the present state of our knowledge, we may answer, that the urea is formed in the blood, and that it is produced from materials that have become effete, the detritus of tissues, as well as from unservicable and superfluous nitrogenous substances in the blood. No animal tissue presents such vital activity, is so much used, and so rapidly worn out, as muscular tissue; it is in this tissue that the metamorphosis of matter proceeds most rapidly and abundantly, and yet, in the large quantities of muscular fluid on which Liebig worked, he could detect no trace of urea, although he found substances from which he could produce urea artificially. We must therefore assume that these substances, as creatine and probably inosic acid, are decomposed in the blood, by the action of the alkalies and of free oxygen, into urea and other matters to be excreted. Moreover, my experiments showing that the superfluous nitrogenous food which enters the blood, and the fact that caffeine, glycine (Horsford), uric acid, and alloxantin (Wöhler and Frerichs),<sup>4</sup> soon after they have

<sup>1</sup> Ann. de Chim. et de Phys. T. 23, p. 90.

<sup>2</sup> Pogg. Ann. Bd. 31, S. 303.

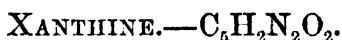
<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 337-8.

<sup>3</sup> Journ. f. pr. Ch. Bd. 11, S. 149.

been taken, perceptibly increase the amount of urea in the urine, support the view that urea is formed in the blood. It is impossible to suppose that this nitrogenous food is first converted into tissue, and subsequently into urea, &c., for we cannot think that a process occurs here, analogous to that exhibited by the percussion-apparatus of Physicists, where a certain number of parts, effecting a percussion, give rise to the repulsion of an equal number of parts. Hence the conversion of this matter can occur in no other place than in the circulating blood, and therefore it is here that the urea must be formed.

That the urea is formed from nitrogenous matters could not be doubted, even if it did not contain nitrogen (and that in so large a quantity); for it is especially after the use of highly nitrogenous food that we find an augmentation of its quantity in the urine. If, however, we should further inquire—from what substances is it produced, and what tissues principally contribute to its formation? we could not, in the present state of our knowledge, give any satisfactory answers to these questions. All that we know is, that urea is a very general product of the decomposition of nitrogenous matters, both naturally within the animal body, and artificially in the laboratory of the chemist. We have already said enough to show that urea is so common a product of the decomposition of nitrogenous bodies, that we could hardly any longer enumerate it among true organic substances, if we tried to establish a distinction between organic and inorganic matter. Moreover, when we treat of uric acid we shall show that, in all probability, a great part of the urea separated by the kidneys from the blood is the product of the decomposition of that acid.

What is the importance of urea in the fluids of the eye, and whether it has any importance, are questions which, at present, cannot be answered.



#### *Chemical Relations.*

*Properties.*—This body, which has also been named *uric oxide* and *urous acid*, occurs, when freshly precipitated, as a white powder, which is neither crystalline nor gelatinous; when dried, it forms pale, yellowish, hard masses, which, on being rubbed, assume a waxy brightness: it is very slightly soluble in water, is insoluble in alcohol and ether, has no action on vegetable colors, and when heated, becomes decomposed without undergoing fusion, developing much hydrocyanic acid and a very peculiar odor, but yielding no urea. It dissolves with considerable facility in ammonia, but on evaporation it loses the greater part of the ammonia, and separates into a yellowish foliaceous mass. It dissolves freely in the caustic fixed alkalies, from which, however, carbonic acid will separate it; it dissolves also in nitric acid without the development of gas, and in sulphuric acid, to which it communicates a yellowish color; it is all but insoluble in hydrochloric and oxalic acids. It does not combine in definite proportions with acids, alkalies, or salts.

*Composition.*—As, from the want of definite combinations, the atomic

weight of this body cannot be ascertained, we can only give the empirical formula, which expresses the simplest relation of the elements in xanthine. This substance was analyzed many years ago by Liebig and Wöhler,<sup>1</sup> and recently by Bodo Unger,<sup>2</sup> with similar results:

Carbon,	.	.	.	.	.	5 atoms,	.	.	.	39.47
Hydrogen,	.	.	.	.	.	2	"	.	.	2.63
Nitrogen,	.	.	.	.	.	2	"	.	.	36.84
Oxygen,	.	.	.	.	.	2	"	.	.	21.06
										<hr/> 100.00

This body has been regarded as uric acid ( $C_5H_2N_2O_3$ ) in a lower state of oxidation; but till some of its compounds or products of decomposition are analyzed, scarcely an hypothesis can be suggested regarding its theoretical constitution.

This body is only classified here with the animal bases, amongst which it cannot properly be reckoned, because, in its elementary composition, it presents much similarity with them, and in a physiological point of view, it approximates to urica, guanine, and cystine.

*Preparation.*—Urinary calculi, in which this body occurs, are dissolved in a solution of potash, and the xanthine is precipitated from the filtered fluid by carbonic acid.

*Tests.*—From the circumstances under which it occurs, this body can only be confounded with uric acid or cystine; under the microscope it may, however, be readily distinguished from them by its amorphous condition. It differs chemically from uric acid, firstly, in its ready solubility in ammonia (hence it is not precipitated from its potash-solution, like uric acid, by hydrochlorate of ammonia); secondly, in its being separated from its potash-solution by carbonic acid, as a precipitate, free from the alkali; thirdly, in its dissolving in nitric acid without effervescence, and on evaporation, leaving a (not red, but) yellow mass, which does not become red on the addition of ammonia. It differs from cystine, not only in its amorphism, but also in its insolubility in hydrochloric and oxalic acids.

### *Physiological Relations.*

*Occurrence.*—This body was discovered in a urinary calculus by Marcet, who, from its behavior with nitric acid, gave it the name of *xanthic oxide*. It has only been found once since, by Stromeyer, in a large calculus removed from a child; and it was from this source that both Liebig and Wöhler, and Unger, obtained the materials for their analyses. Jackson<sup>3</sup> thought that he had found it in a specimen of diabetic urine, but his experiments do not prove that he actually met with this substance. Although I have repeatedly sought for it, I have never been able to find xanthine in diabetic urine; indeed it has never been found in any specimen of urine.

Strahl and Lieberkühn<sup>4</sup> believe that they have discovered xanthine in

<sup>1</sup> Pogg. Ann. Bd. 41, S. 393.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 18.

<sup>3</sup> Arch. d. Pharm. Bd. 11, S. 182.

<sup>4</sup> Harnsaure im Blut u. s. w. Berlin, 1848, S. 112 ff.

human urine, but from the reactions which they describe, the substance in question appears to have been guanine.

[Dr. Davy<sup>1</sup> believes that the urinary secretion of scorpions and spiders consists for the most part of xanthine. The substance he has discovered is doubtless the same as that which Gorup-Besanez and F. Will have regarded as guanine. See p. 160.—G. E. D.]

*Origin.*—So little is known of this substance in reference either to its chemical nature, or its occurrence in the animal body, that we cannot offer any conjecture regarding its genesis.

Many attempts have been made to convert uric acid into xanthine, but they have all been unsuccessful.

Chevallier and Lassaigue<sup>2</sup> have extracted a substance to which they have given the name *xanthocystine*, from the miliary tubercles in a dead body that had been buried for two months. It was insoluble in water and alcohol, but dissolved in ammonia and in the mineral acids; the ammoniacal solution deposited minute white granules on evaporation; hexagonal tablets separated from the acid solutions on evaporation; the substance did not fuse on heating, but puffed up, became yellow and black, and developed an odor of burned horn, and gave off alkaline vapors. The investigation of this substance was not carried any further.

### HYPOXANTHINE.

[Scherer<sup>3</sup> has discovered the occurrence of a white, crystalline, pulverulent substance in the spleen, and in the heart of man and the ox. On analysis it yielded:

Carbon, . . . . .	44.257
Hydrogen, . . . . .	3.219
Nitrogen, . . . . .	40.820
Oxygen, . . . . .	11.704
	<hr/>
	100.000

Its formula is  $C_5H_2N_2O$ . Hence it is xanthine *minus* 1 equivalent of oxygen. Scherer has given it the name of *hypoxanthine*.—G. E. D.]

The following is the best method of preparing hypoxanthine. The fluid obtained by boiling the spleen with water is precipitated by baryta-water; the filtered fluid deposits baryta-salts on evaporation, and must be refiltered and the baryta precipitated by sulphuric acid; all these baryta-precipitates contain hypoxanthine mixed with the phosphate, carbonate, and sulphate of baryta. It is extracted from them by a dilute solution of potash, and is precipitated from this solution, together with uric acid, by hydrochloric or carbonic acid. The hypoxanthine may be obtained in a separate state by dissolving the precipitate in potash, and throwing down the uric acid by hydrochlorate of ammonia.

This substance has been found by Gerhard<sup>4</sup> (one of Scherer's pupils)

<sup>1</sup> Edin. New Phil. Journ. Vol. 40, p. 338, and Vol. 44, p. 125.

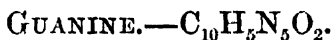
<sup>2</sup> Journ. de Chim. Méd. 3 Ser. T. 7, p. 298. <sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 73, S. 328.

<sup>4</sup> Verh. d. phys.-med. Ges. zu Würzburg. Bd. 2, S. 299.

in the blood of the ox, and by Scherer<sup>1</sup> himself in larger quantity in the blood in leucæmia.

We must know more about the occurrence of hypoxanthine, and its chemical constitution must be further studied, before we can venture to form a judgment, or even to offer an opinion, regarding its physiological value.

[We may take this opportunity of mentioning that Scherer<sup>2</sup> has also found another body in the fluid of the spleen, to which he has given the name of *lienine*; it is crystalline, and according to Scherer's analysis contains no sulphur, but consists of C 53·71, H 8·95, N 4·82, and O 32·52. —G. E. D.]



#### *Chemical Relations.*

*Properties.*—This body is a yellowish-white crystalline powder, devoid of odor or taste, which can bear a temperature of 220° without loss of weight, is insoluble in water, alcohol, and ether, has no action on vegetable colors, and dissolves freely in hydrochloric acid and caustic soda; it unites with acids, forming unstable salts; on mixing its sulphate with a very large quantity of water, there is a separation of the hydrate of guanine, which does not lose its combined water till it is raised to a temperature of 100°.

*Composition.*—This body was discovered by Bodo Unger;<sup>3</sup> it was at first mistaken for xanthine, but subsequently, by analysis of the free body and its salts, it was ascertained to be a distinct, weak base. According to the formula deduced from his analyses, it consists of:

Carbon,	.	.	.	.	10 atoms,	.	.	.	39·73
Hydrogen,	.	.	.	.	5 "	.	.	.	3·31
Nitrogen,	.	.	.	.	5 "	.	.	.	46·36
Oxygen,	.	.	.	.	2 "	.	.	.	10·60
									<hr/> 100 00

Its atomic weight = 1887·5. The hydrate consists, according to Unger, of 2 atoms of water and 3 atoms of guanine. On account of its basic nature, Berzelius<sup>4</sup> regards it as ammonia with a nitrogenous adjunct =  $\text{H}_3\text{N} \cdot \text{C}_{10}\text{H}_5\text{N}_4\text{O}_2$ .

*Combinations.*—Like caffeine and theobromine, and other weak bases, guanine readily unites in several proportions with acids, but, like the above-named substances, parts with them readily on the addition of large quantities of water, so that the pure base, mostly as a hydrate, is separated, while an acid salt remains in solution.

*Hydrochlorate of guanine:* the neutral salt,  $3(\text{C}_{10}\text{H}_5\text{N}_5\text{O}_2 \cdot \text{HCl}) + 7\text{H}_2\text{O}$ , crystallizes in bright yellow needles, loses all its water under 100°, and

<sup>1</sup> Verh. d. phys.-med. Ges. zu Würzburg. p. 323.

<sup>2</sup> Ibid. p. 298.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 51, S. 395 ff, and Bd. 58, S. 28–31; Pogg. Ann. Bd. 65, S. 222–239, and Ann. d. Ch. u. Pharm. Bd. 59, S. 58–73.

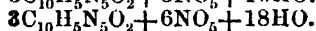
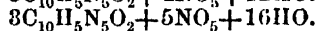
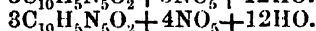
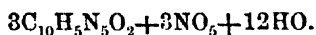
<sup>4</sup> Jahresber. Bd. 27, S. 678.



all its hydrochloric acid above that temperature: the acid salt,  $C_{10}H_5N_5O_2 + 2HCl$ , loses half its hydrochloric acid at a moderate temperature: with bichloride of platinum it forms a crystalline compound,  $C_{10}H_5N_5O_2 \cdot HCl + PtCl_2 + 4HO$ , which is as insoluble in cold water as the ammoniochloride of platinum, but dissolves very freely in hot water. The following basic hydrochlorate has also been obtained:  $2C_{10}H_5N_5O_2 + HCl$ .

*Sulphate of guanine*,  $C_{10}H_5N_5O_2 \cdot HO \cdot SO_3 + 2HO$ , crystallizes in yellow needles, often an inch in length.

*Nitrate of guanine* was obtained by Unger in several proportions:



The phosphate, oxalate, and tartrate of guanine may also be obtained.

*Guanine-soda*,  $C_{10}H_5N_5O_2 + 2NaO + 6HO$ , is precipitated from the soda-solution on the addition of alcohol: it is a foliaceous crystalline mass, which attracts carbonic acid from the air, and effloresces. At  $100^\circ$  it loses all its water; on the addition of water one portion of the guanine separates, and another portion remains in solution with an excess of soda. Guanine also unites with certain salts, as, for instance, with nitrate of silver, forming crystalline compounds.

*Products of its metamorphosis.*—*Guanic acid*,  $C_{10}H_3N_4O_7$  (termed *hyperuric acid* by Unger), is obtained by digesting for 24 hours, at a temperature of  $125^\circ$ , 3 parts of guanine, 5 of chlorate of potash, 5 of water, and 30 of hydrochloric acid; it crystallizes in short rhombic prisms with oblique terminal surfaces, is devoid of color, odor, and taste, reddens moistened litmus, is slightly soluble in water and in acids, but dissolves freely in the caustic alkalies and their carbonates, and on dry distillation yields hydrated cyanic acid, together with water and carbon.

*Preparation.*—Guanine was obtained by Unger from guano, which he digests with diluted milk of lime till the fluid, when boiled, no longer appears brown, but assumes a faint greenish-yellow color; it is then filtered and treated with hydrochloric acid; in the course of a few hours the guanine, with a little uric acid, separates; the sediment is then dissolved in hydrochloric acid, from which it is deposited in a crystalline form as a hydrochlorate; from this the guanine is finally separated by ammonia.

*Tests.*—Guanine is especially to be distinguished both from xanthine and from uric acid by its forming distinctly crystallizable salts with acids. Moreover, the difference of its behavior with nitric acid is quite sufficient to prevent it from being mistaken for uric acid.

### *Physiological Relations.*

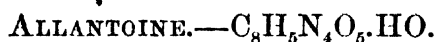
*Occurrence.*—Unger has, as we have already mentioned, found guanine in guano (the excrements of certain sea-fowls); it has recently also been found in the excrements of spiders by F. Will and Gorup-Besanez,<sup>1</sup> who

<sup>1</sup> Gelehrte Anz. d. k. bair. Ak. d. Wiss. 1848, S. 825-828; [and more fully in a memoir "on guanine as an essential constituent of certain secretions of the invertebrata," in Ann. d. Ch. u. Pharm. Bd. 69, S. 117.—G. E. D.]

think it very probable that this substance occurs in the green organ of the river craw-fish, and in the Bojanian organ in the fresh-water mussel.

If the constant occurrence of this substance in the urine, which Strahl and Lieberkühn<sup>1</sup> regarded as xanthine (but which, from its solubility in hydrochloric acid, would rather seem to be guanine), be confirmed by further investigations, we should have to classify guanine among the general products of excretion of the animal organism.

*Origin.*—From everything connected with the occurrence of guanine there can be no doubt that, like the nitrogenous compounds to which it is allied, it is a product of the metamorphosis of the nitrogenous matters of the animal body. Nothing is, however, known, on which we can even hazard a conjecture regarding the conditions under which it is formed.



#### *Chemical Relations.*

*Properties.*—This body forms colorless, hard prisms, of the rhombic primitive form, which have a strong vitreous brilliance; it is devoid of smell and taste, dissolves in 160 parts of cold water, and more easily in hot water; it crystallizes from its hot alcoholic solution, is insoluble in ether, is unaffected by exposure to the atmosphere, does not redden litmus, and chars, when heated, without fusing. It dissolves in solutions of the caustic alkalis and their carbonates, when these are warmed, but crystallizes from them in an unchanged condition as they cool; it is decomposed by concentrated caustic alkalis, taking up water and resolving itself into oxalic acid and ammonia ( $\text{C}_8\text{H}_5\text{N}_4\text{O}_5 + 7\text{HO} = 4\text{H}_3\text{N} + \text{C}_2\text{O}_3$ ); when boiled with concentrated sulphuric acid, it also takes up water, developing carbonic acid and carbonic oxide, and leaving sulphate of ammonia. On warming it with nitric acid (of 1·2 to 1·4 specific gravity), it becomes decomposed into urea and allantoic acid (3 atoms of allantoin, taking up 7 atoms of water, yield 2 atoms of urea and 2 atoms of allantoic acid, for  $\text{C}_{24}\text{H}_{15}\text{N}_{12}\text{O}_5 + 7\text{HO} = \text{C}_4\text{H}_8\text{N}_4\text{O}_4 + \text{C}_{20}\text{H}_{14}\text{N}_8\text{O}_{18}$ ).

Allantoin enters into combination with the oxides of lead and silver.

*Composition.*—Liebig and Wöhler<sup>2</sup> were the first who accurately determined\* the composition of crystallized allantoin, and they deduced the above formula from its silver-compound, according to which it consists of:

Carbon,	.	.	.	.	8 atoms,	.	.	.	.	30·38
Hydrogen,	.	.	.	.	5 "	.	.	.	.	8·16
Nitrogen,	.	.	.	.	4 "	.	.	.	.	35·44
Oxygen,	.	.	.	.	5 "	.	.	.	.	25·32
Water,	.	.	.	.	1 "	.	.	.	.	5·70
										<hr/> 100·00

The atomic weight of the hypothetical dry allantoin = 1862·5.

This body cannot be reckoned amongst the organic bases, since it does

<sup>1</sup> Op. cit.

<sup>2</sup> Pogg. Ann. Bd. 31, S. 561.

not combine with any acid; but from the analogy of its composition, and the circumstance that we cannot find a more appropriate position for it than amongst the nitrogenous products of the metamorphosis of animal matters, we deemed it best to insert it in this place. No rational formula can be assigned for it; we may, however, remark, that it exactly contains the elements of 4 atoms of cyanogen and 5 atoms of water.

*Combinations.*—The *silver-compound*,  $C_8H_5N_4O_5 \cdot AgO$ , is obtained by mixing nitrate of silver with a boiling saturated solution of allantoine, and then adding ammonia as long as a precipitate continues to be produced: it forms a white powder which, when examined microscopically, is found to consist of clear, perfectly spherical particles.

The *lead-compound* is obtained, on boiling an aqueous solution of allantoine with oxide of lead; it is crystallizable.

*Products of its metamorphosis.*—*Allantoic acid*,  $C_{10}H_7N_4O_9$ , which is obtained in the manner we have already described, occurs as a tough, amorphous, white mass, soluble in water, but insoluble in alcohol and ether, and forms soluble salts with the alkalis and earths (Pelouze).<sup>1</sup> Attention has been drawn to the fact that this acid contains exactly 3 atoms of water more than uric acid under the older formula ( $C_{10}H_4N_4O_6 + 3HO = C_{10}H_7N_4O_9$ ).

*Preparation.*—On evaporating the allantoic fluid of the fœtus of a cow or the urine of a young calf to a thin syrup, without permitting it to boil, and then allowing it to stand for a few days, we obtain crystals of allantoine mixed with phosphate and urate of magnesia; by stirring it with cold water and decanting, most of the viscid matter, consisting of urate of magnesia, is removed, while the crystals of allantoine and phosphate of magnesia rapidly sink to the bottom; hot water extracts the allantoine, leaving the magnesian salt undissolved; the solution of allantoine is then decolorized with animal charcoal, and evaporated till it recrystallizes.

Allantoine may also be obtained artificially from uric acid (see “Uric Acid”) by boiling it with peroxide of lead, the products of decomposition being oxalate of lead, urea, and allantoine; when the boiling fluid has been freed by filtration from oxalate of lead, and allowed to cool, the allantoine separates in crystals.

*Tests.*—This body can only be recognized with certainty by an accurate determination of its crystalline form, or by an elementary analysis either of itself or its silver-compound.

### *Physiological Relations.*

*Occurrence.*—Vauquelin and Buniva thought that they had found allantoine in the Liquor Amnii of a cow, but Lassaigne<sup>2</sup> proved that it is peculiar to the Liquor Allantoidis. It has recently been found by Wöhler<sup>3</sup> in considerable quantity, in the urine of young calves. It has as yet been found nowhere else in the animal organism.

According to Wöhler, the allantoine from calves' urine presents the

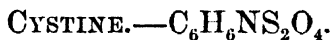
<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. T. 6, p. 69.

<sup>2</sup> Ibid. T. 17, p. 301.

<sup>3</sup> Nachrichten der k. Gesellsch. d. Wiss. zu Göttingen, 1849. No. 5, S. 61–64; [and more fully in Ann. d. Ch. u. Pharm. Bd 70, S. 229.—a. e. d.]

peculiarity that it differs in the character of its crystals from that which is obtained from the allantoic fluid or from uric acid; the crystals grow together in bundles, and their terminal surfaces are no longer distinct, while pure allantoin appears in isolated well-formed prisms. This difference, however, only depends on the admixture of a foreign substance, whose quantity is much too minute to produce any appreciable influence on the result of its elementary analysis. By combining it with oxide of silver, and then decomposing the compound, we obtain it in as pure and isolated a state as when we prepare it from the allantoic fluid or from uric acid.

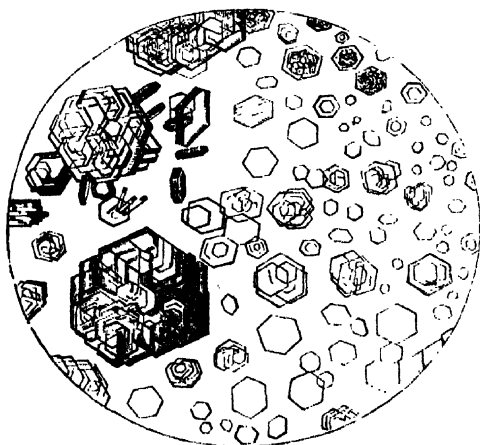
*Origin.*—That allantoin is a product of the metamorphosis of nitrogenous food or of tissue in the animal organism, is sufficiently obvious from the circumstances under which it occurs, but any nearer indication of the chemical process on which its formation depends is impossible, since we have no idea of its rational composition. The two following facts may, however, probably indicate the way in which its formation may at some future time be explained: firstly, it only occurs in the urine of the fœtus and of recently-born animals, and disappears after the use of vegetable food; secondly, as has been discovered by Wöhler, it occurs in the urine of sucking calves, together with uric acid and urea, but without hippuric acid; hence the idea suggests itself that allantoin and hippuric acid exclude or stand in the place of one another, which might rather have been expected of uric acid, from which allantoin may be artificially prepared.



*Chemical Relations.*

*Properties.*—This body occurs in colorless, transparent, hexagonal plates or prisms, is devoid of taste and smell, and is insoluble in water

Fig. 9.



Cystine from urinary calculus, and recrystallized from ammonia.

and alcohol; it dissolves in oxalic acid and in the mineral acids, forming with them saline combinations, most of which are crystallizable, but it

does not unite with acetic, tartaric, or citric acid: it is decomposed by nitric acid, leaving, on the evaporation of the fluid, a reddish-brown mass; it dissolves freely in the caustic fixed alkalis and their carbonates. It dissolves in caustic ammonia, but does not unite with it, so that on evaporation it crystallizes unchanged. It is insoluble in carbonate of ammonia; hence it is best precipitated from its acid solutions by carbonate of ammonia, and from its alkaline solutions by acetic acid.

Cystine does not fuse on the application of heat, but it burns with a bluish-green flame, developing at the same time a very peculiar acid odor; on dry distillation it develops a stinking empyreuma and ammonia, and leaves a voluminous porous coal. On boiling it with alkalis, ammonia is first developed, and subsequently an easily inflammable gas, which burns with a blue flame.

*Composition.*—Cystine has been analyzed by Prout, Baudrimont, Thaulow,<sup>1</sup> and Marchand,<sup>2</sup> with perfectly identical results, yielding the above formula, according to which this substance contains:

Carbon, . . . . .	6 atoms,	. . . . .	30.000
Hydrogen, . . . . .	6 “	. . . . .	5.000
Nitrogen, . . . . .	1 “	. . . . .	11.666
Sulphur, . . . . .	2 “	. . . . .	26.667
Oxygen, . . . . .	4 “	. . . . .	26.667
			<hr/>
			100.000

Its atomic weight = 1336.0.

Since cystine, which has also received the name of *cystic oxide*, unites with certain acids to form crystalline salts, Berzelius classifies this body with the combinations of conjugated ammonia  $\text{H}_3\text{N} \cdot \text{C}_6\text{H}_3\text{S}_2\text{O}_4$ . If, however, this view be correct, much is still wanting for the establishment of the rational formula of cystine, for the most important question regarding its constitution still remains unexplained, namely, in which form or combination the sulphur is contained, in the cystine or in this adjunct. The chemical investigations regarding cystine, which have been hitherto instituted, do not tend to support any hypothesis.

*Combinations.*—*Hydrochlorate of cystine*,  $\text{C}_6\text{H}_5\text{NS}_2\text{O}_4 \cdot \text{HCl}$ , crystallizes without water in plates grouped in a star-like form. Berzelius<sup>3</sup> obtained the combination with bichloride of platinum by direct union; this salt is not crystallizable; it dissolves easily in water and alcohol, but is insoluble in ether.

*Nitrate of cystine*,  $\text{C}_6\text{H}_6\text{NS}_2\text{O}_4 \cdot \text{HO} \cdot \text{NO}_5 + \text{HO}$ , crystallizes readily, losing its one atom of water at  $85^\circ$ .

*Preparation.*—Urinary calculi, in which cystine occurs, are dissolved in a solution of potash, and the cystine is precipitated from this solution by acetic acid; or we dissolve them in ammonia, and allow the filtered fluid to evaporate in the air.

*Tests.*—Cystine is characterized by the readiness with which it crystallizes in well-formed hexagonal plates, which may be distinguished with great ease under the microscope, and by its solubility both in alkalis and mineral acids. Further, it may be known by the peculiar

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 27, S. 197.

<sup>2</sup> Journ. f. pr. Ch. Bd. 10, S. 15–18.

<sup>3</sup> Jahresber. Bd. 27, S. 631.

odor which it develops on dry distillation and on burning, which is unlike that evolved by any other similar substance. Liebig has given the following test for cystine. The potash-extract of the substance in which we are searching for cystine must be decomposed with a solution of oxide of lead in caustic potash; if, on the application of heat, there be a precipitation of sulphide of lead, cystine is probably present; we must, however, previously satisfy ourselves that no other sulphurous body, as, for instance, mucus, albumen, &c., be simultaneously present.

If cystine be mixed with a small quantity of the urates, the two substances may be separated by the aid of boiling water, in which the former is insoluble. Uric acid occasionally appears under the microscope in the form of hexagonal tablets, but we should never trust in these cases to microscopic examination alone.

### *Physiological Relations.*

*Occurrence.*—Cystine was originally discovered by Wollaston,<sup>1</sup> in a urinary calculus. Calculi of this nature, although very rare, have since been found by many other chemists, as, for instance, Prout, Taylor, Baudrimont, Lassaigne, Dranty, Civiale, Buchner, and Bird. Bird<sup>2</sup> and Mandl<sup>3</sup> remark that they have often found cystine dissolved in the urine, from which Bird precipitates it by acetic acid; it also occurs as a sediment mixed with urate of soda. The pathological process accompanying the appearance of cystine in the urine is altogether unknown. Bird thinks there is some connection between it and the scrofulous diathesis; others fancy that they see connection between cystine and diabetes; but none of these conjectures are supported by the results of experience. In the examination of 129 urinary calculi, Taylor found only two that contained cystine. This substance has been found nowhere but in the urine.

*Origin.*—As no other urinary constituent contains sulphur,<sup>4</sup> the occurrence of this highly sulphurous body in the urine is the more singular, and we should consequently expect that some essential alteration of the chemico-vital processes must have taken place before this substance could be produced, but all that we learn from the simultaneous morbid phenomena completely disappoints us in the assumption that the excretion of cystine must probably be preceded by a certain group of symptoms, from which something might be concluded regarding the production of this body. Taurine is the only other body with which we are acquainted that is equally rich in sulphur; no other animal bodies in which sulphur occurs, as albumen, casein, fibrin, &c., contain at most more than 2%, while in this substance there is 25%. Hence, in a chemical point of view, a connection might be suspected between taurine and cystine, and the rational physician should consequently direct his attention to the manner in which the functions of the liver are performed, whenever cystine presents itself in the urine.

<sup>1</sup> Phil. Trans. 1810, p. 223.

<sup>2</sup> Urinary Deposits, &c. 3d Edition, p. 188.

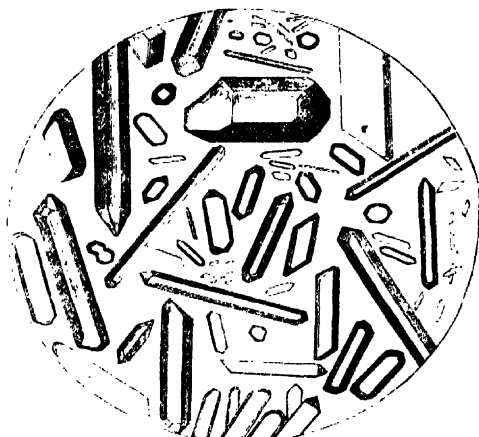
<sup>3</sup> Journ de Chim. Méd. 1838, p. 355.

<sup>4</sup> [This statement is too general. Dr. Ronalds has shown that the extractive matters of the urine contain an unknown sulphur-compound. See Phil. Trans. 1846, p. 461.—G. E. D.]

TAURINE.— $C_4H_7NS_2O_6$ .

*Properties.*—This substance, which was formerly termed *biliary asparagin*, crystallizes in colorless, regular hexagonal prisms with four and six-sided sharp extremities (the elementary form is that of a right rhombic prism, the angles formed by the edges of the sides being  $111^{\circ}44$  and  $68^{\circ}16$ ) (Fig. 10); it is hard, crunches between the teeth, has a cool-

Fig. 10.



Taurine from Ox-gall, crystallized from hot water.

ing taste, resists the action of the atmosphere, dissolves in 15.5 parts of water, and in 573 of spirit of wine (of 0.835 specific gravity), but is insoluble in anhydrous alcohol and ether, and has no action on vegetable colors. It dissolves, without undergoing change, even at the boiling-point, in the mineral acids, but forms no compounds with them. It is not precipitated from its solution either by tannic acid or by the metallic salts. On heating, it fuses, puffs up, and develops much acetate of ammonia, and a thick brown oil; if it be inflamed in the air, it develops much sulphurous acid; if it be dissolved in caustic potash, and the solution boiled down till it becomes thick, it develops pure ammonia gas, and leaves a residue consisting solely of sulphite and acetate of potash. The sulphur in taurine cannot be detected in the moist way either by nitric acid or by aqua regia.

*Composition.*—Taurine was first discovered by Gmelin in the bile, and was soon afterwards analyzed with very similar results by Demarçay, Pelouze, and Dumas; these chemists, however, entirely overlooked the existence of sulphur in this body, the discovery of which was reserved for Redtenbacher,<sup>1</sup> from whose analyses it was found to consist of:

Carbon,	.	.	.	.	4 atoms,	.	.	.	19.20
Hydrogen,	.	.	.	.	7 "	.	.	.	5.60
Nitrogen,	.	.	.	.	1 "	.	.	.	11.20
Sulphur,	.	.	.	.	2 "	.	.	.	25.60
Oxygen,	.	.	.	.	6 "	.	.	.	38.40
									<hr/> 100.00

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd 57, S. 170-174.

As this body has not yet been combined with any other in a definite proportion, its atomic weight cannot be determined with accuracy; but it must not be reckoned among the bases, and we are still perfectly in the dark regarding its rational composition. Redtenbacher<sup>1</sup> attempted to elucidate this point; finding that by the action of potash, taurine was decomposed into ammonia, acetic acid, and sulphurous acid, he was somewhat inclined to believe that taurine is a combination of sulphurous acid with aldehyde and ammonia (since  $2\text{SO}_2 + \text{H}_3\text{N} + \text{C}_4\text{H}_4\text{O}_2 = \text{C}_4\text{H}_7\text{NS}_2\text{O}_6$ ), and that it might probably be artificially prepared from these substances, as urea is obtained from cyanate of ammonia. Indeed, on passing sulphurous acid into an alcoholic solution of aldehyde-ammonia he obtained a white crystalline body isomeric with taurine; it is, however, not identical with taurine, but must be regarded as an acid sulphite of aldehyde-ammonia; it reddens litmus, gradually changes on exposure to the air, turns yellow at  $100^\circ$ , and at a higher temperature becomes brown, and finally develops an odor resembling that of burned taurine. Hence, notwithstanding these ingenious experiments of Redtenbacher's, the rational constitution of taurine remains still unexplained.

[Strecker<sup>2</sup> has recently succeeded in forming taurine artificially from isethionate of ammonia,  $\text{NH}_4\text{O} \cdot \text{C}_4\text{H}_5\text{O} \cdot 2\text{SO}_3$ , which  $= \text{C}_4\text{H}_7\text{NO}_6\text{S}_2 + 2\text{H}_2\text{O}$ , and therefore, only differs from taurine by two equivalents of water. This salt fuses at  $120^\circ\text{C}$  without disengaging ammonia, and Scherer hoped that at a still higher temperature it would lose water. He first found that taurine might be heated to  $240^\circ\text{C}$ , without decomposition or fusion; and he then heated isethionate of ammonia to  $236^\circ\text{C}$ , and kept it at this temperature till it had lost 11% of weight. The mass was dissolved in water; on the addition of alcohol it was precipitated in crystals; this precipitate, dissolved in water, furnished by spontaneous evaporation large crystals exactly identical with the crystals of taurine obtained from bile. Like taurine, they bear exposure to  $240^\circ$ , without fusing or acquiring color; they evolve no ammonia with a solution of potash; they do not precipitate the salts of baryta when boiled with nitric or nitro-muriatic acid; when fused with potash and nitrate of potash, they evolve ammonia, and the mass contains sulphuric acid. All these properties being the same as those of taurine, and its mode of formation proving that its composition is similar, this product is identical with the taurine of the bile.—G. E. D.]

*Preparation.*—Taurine is usually obtained from ox-gall. The bile, freed from its mucus by an acid, or its alcoholic extract, is mixed with hydrochloric acid, and boiled for some hours, till the cholidic acid is completely formed from the nitrogenous acids of the bile; the acid fluid, after the removal of the cholidic acid by filtration is rapidly evaporated, causing the chloride of sodium to crystallize; the acid mother-liquid is then treated with five or six times its bulk of boiling alcohol, from which, as it cools, the taurine separates in needles; by recrystallization in water, it is obtained in a state of purity.

*Tests.*—Taurine may be distinguished from every other substance by its crystalline form (which, under the microscope, is as distinct in small crystals as in large ones), by its property of developing sulphurous acid

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 37-45.

<sup>2</sup> Compt. rend. T. 39, p. 63.



when heated in a glass tube open at both ends, or on a platinum spatula, and finally, by the circumstance that when boiled with caustic potash, it does not form a black solution, but develops ammonia, and leaves a residue consisting solely of sulphurous and acetic acids in combination with potash.

### *Physiological Relations.*

*Occurrence.*—Taurine has never been found isolated in the healthy organism; it appears to be contained preformed in normal bile, and to occur there as an adjunct of the already described cholic acid; at all events, it only occurs in an isolated state in decomposed or morbid bile. After the removal of the mucus, the only sulphur-compound, in those animals in which the bile contains sulphur, is taurine conjugated with cholic acid. At the present time we know, by the researches of Bensch,<sup>1</sup> that sulphur exists in the bile of the ox, the sheep, the fox, the bear, the dog, the wolf, the goat, the domestic hen, and certain fresh-water fish; and Schlieper<sup>2</sup> has found it most abundant in the bile of serpents. From the bile of the pig, Strecker and Gundelach<sup>3</sup> were unable to obtain taurine, and they found no sulphur in it, although Bensch had detected a small quantity. Doubts have been expressed whether sulphur, and consequently taurocholic acid, exists in human bile, but Gorup-Besanez<sup>4</sup> has so completely set this point at rest, that my evidence founded on the crystallometric determination of taurine artificially obtained from human bile is superfluous. In diseased bile taken from the dead body taurine is especially found, when, as is sometimes the case, the bile has an acid reaction; thus Gorup-Besanez found taurine in the bile of a person who had died from arachnitis.

Although some of the products of the decomposition of bile occur in the excrements, especially in cases of diarrhoea, taurine has never yet been found there: neither has it been detected in bilious urine.

*Origin.*—If we consider that the excreted products of the animal organism are usually highly oxidized organic matters, and that most of the matters separated from the blood and even deposited in the tissues, differ from the food in containing a larger amount of oxygen, it must at first sight strike us as singular that a substance so rich in sulphur as taurine, either alone or in combination, should be produced, even in the normal state of the body, from the animal fluids, which are almost universally saturated with free oxygen. Although Redtenbacher failed in obtaining taurine artificially, his admirable researches render it highly probable that the sulphur in taurine exists in an oxidized state, as indeed may be inferred, from the fact that it cannot be recognized in this substance by means of the ordinary fluid oxidizing agents. The genesis of taurine should therefore not be sought in a deoxidizing process in the blood (a very improbable process), but rather in a process of oxidation. If, however, taurine be the product of an oxidation, the source of its formation should hardly be sought in the liver, since the blood that is poorest in oxygen is supplied to this organ. This simple induction leads

<sup>1</sup> Ann. d. Ch. u Pharm. Bd. 65, S. 194–203.

<sup>2</sup> Ibid. Bd. 60, S. 109–112.

<sup>3</sup> Ibid. Bd. 62, S. 205–232.

<sup>4</sup> Unters. üb. Galle. Erlangen, 1846. S. 31–37.

us to refer the seat of the formation of taurine, or at least of its proximate constituents, to the blood, where, however, it cannot be detected for the same reason that so long prevented the presence of urea from being ascertained. Nothing is at present known regarding the different steps that occur in the formation of taurine; it is, however, not improbable that the sulphur of the albuminous food in its conversion into the elements of tissues, which are either free from or poor in sulphur, yields in part the materials for the formation of taurine.

*Uses.*—If we can conjecture with some probability regarding the origin of taurine, we are even less fortunate in reference to the function which the taurine excreted with the bile in the intestine, exerts in the animal organism, since in this point of view we are entirely devoid of facts on which to hang even a bare induction. No conclusion can be drawn regarding the further use of this substance in the animal body, from the negative fact that hitherto no taurine has been found in normal excrements, since accurate and sufficiently minute experiments have not yet been made on this subject. As there are some animals, as, for instance, the pig, which, although they secrete bile copiously, separate no taurine by the hepatic organs, it appears that at all events it is unimportant to the process of digestion. But that taurine, even if first separated from the blood, should be again resorbed from the intestine into the blood, and being there burned, should serve as a material for supporting the animal heat, appears to us not impossible, but certainly improbable. (See "Taurocholic Acid.")

## CONJUGATED ACIDS.

Although we may not feel justified in directly introducing into physiological chemistry all the transient views which have arisen in theoretical chemistry; and although we would wish to abstain from those more than hypothetical opinions regarding the theoretical constitution of organic bodies, which are forever rising, and as rapidly disappearing; yet we ought not to omit all reference to the present state of theoretical chemistry, but should be ready to appropriate to physiological chemistry, every acquisition which seems likely to be fruitful in results. It would by no means further the progress of physiological chemistry at once to transfer to it all the hypotheses or fictions that may have been advanced in pure chemistry. If we were to attempt to support these chemical hypotheses with others of a physiological nature, the foundation of physiological chemistry would be very unstable, and finally the whole superstructure would be an aerial image of the fancy (and of these images we have already an abundance) rather than an experimental science based on pure induction. It is, however, necessary for the progress of science, that in accordance with the present state of chemical theory we should establish certain general propositions, which not only furnish us with a comprehensive expression for a number of frequently recurring facts,

but guide inquiry in various directions, and finally present us with certain points of support for the due understanding of our scientific material. Amongst these general propositions we reckon the method which is now becoming tolerably common in theoretical chemistry, of considering certain bodies as conjugated or copulated combinations. We shall, however, place no more exclusive dependence on this theory, as it has been carried out by Laurent and Gerhardt,<sup>1</sup> or Strecker,<sup>2</sup> or Kolbe,<sup>3</sup> than on the theory of organic radicals and of electro-chemical dualism of a Berzelius, or on the theory of substitutions and metalepsy of a Dumas. If we even venture on a reference to eclecticism, it must be in the choice of those supports which one branch of science, in its early stage, is compelled to borrow from another. It is only in this point of view that we wish to justify the establishment of the group of conjugated acids in zoo-chemistry.

We have already had occasion to refer to a series of organic acids which, according to the excellent investigations of Kolbe, may be regarded as carbo-hydrogens conjugated with oxalic acid: indeed Kolbe is inclined to regard all the groups of acids we have noticed, which contain 3 atoms of oxygen, as combinations of oxalic acid with carbo-hydrogens. These illustrations are sufficient to indicate the idea which we attach to the expression, *conjugated* or *copulated* acids. We have become acquainted with acids which, in opposition to the ordinary rules of chemistry, not only lose nothing of their acidity, but (which is most singular) perfectly retain their former saturating capacity, when united with another and a more basic body; after being combined with the so-called adjunct (copula), this acid still saturates the same quantity of base as if the organic matter associated with it did not exist; and this dependent—the adjunct—which follows the acid as an integral constituent in all its combinations, exerts an essential influence on its physical and even on many of its chemical properties. Thus, for instance, oxalic acid, which in its ordinary state is so readily decomposed by heat, becomes volatile by its conjugation (accouplement) with the above-named carbo-hydrogens; the stability is, however, most obvious in those acids in which such easily decomposable bodies as hyposulphurous or hyponitric acid are conjugated; their salts being altogether dissimilar from those of the non-conjugated acids in their crystalline form, solubility, amount of water, &c.

In combinations of this kind the electro-chemical polarity is entirely lost; the older dualistic views of chemistry here altogether fail us; we must therefore here assume another ground of chemical attraction than that of opposite polarity, and this view is confirmed by the circumstance that these compounds cannot be decomposed according to our ordinary chemical principles, that is to say, by simple or double elective affinity. They also no more admit of being decomposed into their proximate constituents, that is to say, into the acid and the adjunct, than of being directly formed from them. Most of the conjugated acids are only formed when the adjunct in its nascent state comes in contact with the acid;

<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. T. 24, p. 200-208.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 47-55.

<sup>3</sup> Handwörterb. d. Chemie. Bd. 3, S. 489 444.

and conversely it is only very few of them that can be decomposed into the acid and the adjunct, and even in this case the adjunct invariably assimilates water, and it is impossible to determine with certainty whether the isolated hydrated body in its anhydrous condition actually constituted the adjunct, or whether the latter body was represented by some other group of atoms. This favorable condition, however, very rarely aids us; for generally, in our attempts to separate the adjunct from the acid, the former becomes so decomposed that we can arrive at no conclusion regarding its nature: and this is the reason why chemists, when they enter into the general consideration of the laws of conjugated acids have to trust more or less to hypotheses; and it would scarcely be in accordance with our views to follow their track. We shall, however, be compelled to devote some attention to these hypotheses when we treat of the acids of this class, pertaining to zoo-chemistry; and we will here only remark that we will subsequently treat of those combinations of organic acids with organic oxides in which all acidity has disappeared, and which have been named by Berzelius *haloid salts*, whilst other chemists of the present day have included them in the category of conjugated compounds.

Most of the known conjugated acids are formed by the action of sulphuric or nitric acid on organic substances. in the following group, picric acid is the only one we will consider in any detail, partly by way of general illustration, and partly because it occurs more frequently than the others as a product of the decomposition of different nitrogenous substances by nitric acid. The other acids of this class, to which reference may be made in zoo-chemistry, will be considered under the head of the substances from which they are derived.

There are but few of the pure organic acids whose adjunct can be determined with much probability. It necessarily arises from the nature of these substances, that conjugated organic acids can be decomposed into acids and their adjuncts with much less facility than the conjugated mineral acids, and that their proximate constituents cannot be ascertained without difficulty. We have ventured in the following pages to enumerate nitrogenous organic acids in the group of conjugated acids, not that the composition of each one can with certainty be referred to a nitrogenous adjunct and an acid, but because the study of the products of decomposition of such bodies renders it tolerably evident that all nitrogenous acids, more especially on account of their high atomic weight, are composed of proximate constituents, of which the nitrogenous one scarcely at all contributes to the acidity of the combination.

This, however, is pure conjecture; but, at the same time, in considering the nitrogenous acids, we should have to adopt an arbitrary classification, if we were to consider those in which the conjugate constitution has to any extent been proved, distinct from those in which no evidence of this nature has been obtained. Between these two classes there exist so many analogies that it would be of no practical utility to attempt such a separation.

PICRIC ACID.— $C_{12}H_2N_3O_{13}.HO.$ 

*Properties.*—This acid, which was formerly known as *carbonitric acid*, *carbazotic acid*, and *Welter's bitter*, crystallizes in yellow, glistening plates or prisms, fuses when carefully heated, and admits of being sublimed undecomposed, but when rapidly heated decomposes with explosions; it is devoid of odor, has a very bitter taste, and dissolves slightly in cold and readily in hot water, the solution being of a yellow color; it dissolves freely in alcohol and ether, and reddens litmus; when heated with phosphorus or potassium it decrepitates violently; it is not decomposed by chlorine, nitric or hydrochloric acids, or by *aqua regia*.

*Composition.*—According to the above formula this acid consists of:

Carbon,	.	.	.	.	.	12 atoms,	.	.	.	81.44
Hydrogen,	.	.	.	.	.	2 "	.	.	.	0.87
Nitrogen,	.	.	.	.	.	3 "	.	.	.	18.84
Oxygen,	.	.	.	.	.	13 "	.	.	.	45.42
Water,	.	.	.	.	.	1 "	.	.	.	3.93
										<hr/> 100.00

The atomic weight of the hypothetical anhydrous acid = 2750.0; and its saturating capacity = 3.636. Chemists are not agreed regarding the rational formula of this body; they unite in regarding it as a conjugated nitric acid, but there is much difference of opinion regarding the nature of the adjunct. Berzelius writes this acid as =  $(C_{12}H_2NO_3.NO_5) + NO_5.HO$ , but there is little to support the view of a salt-like adjunct such as is here assumed. We know, for instance, that the group of atoms  $NO_4$  is substituted in aniline and certain other bodies for an equivalent of hydrogen, and it is now pretty generally assumed that such substitutions of more negative matters in the place of hydrogen for the most part only extend to the hydrogen contained in the adjunct; if, therefore, we assign to picric acid only a hypothetical formula, it will at all events not be an irrational one, if we consider that in the adjunct  $C_{12}H_2$ , 2 atoms of hydrogen are replaced by 2 atoms of  $NO_4$ , and write with the acid as =  $C_{12}(H_2.2NO_4).NO_5.HO$ . Laurent regards picric acid, not as a conjugated acid, but as carbonic acid ( $C_{12}H_6O$ ) in which 3 atoms of hydrogen are replaced by 3 atoms of  $NO_4$ , and hence he writes it as =  $C_{12}(H_2.3NO_4)O.HO$ .

*Combinations.*—The *picrates* are crystallizable, yellow, and for the most part soluble in water; when rapidly heated they decrepitate with much violence.

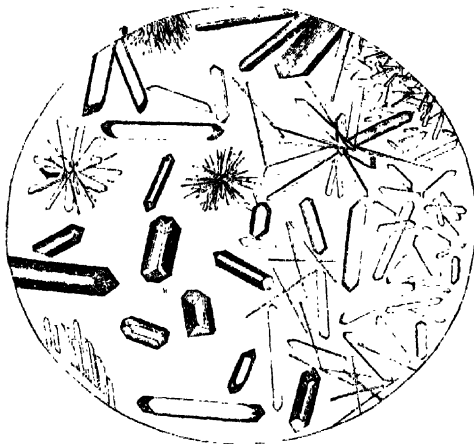
*Picrate of potash* is one of the most insoluble salts of this acid; it crystallizes in long, glistening, yellow, iridescent prisms, and dissolves in 260 parts of cold, and 14 parts of hot water. With alkaline earths and metallic oxides this acid has a tendency to form basic and very insoluble salts.

*Preparation.*—This acid is formed by the action of concentrated nitric acid on many vegetable and animal substances. Thus, for instance, in heating salicin with nitric acid, we obtain crystals of pure picric acid. It is likewise produced in large quantity on decomposing silk with nitric acid; it is, however, most commonly obtained by boiling indigo with nitric acid.

HIPPURIC ACID.— $C_{18}H_8NO_6.HO$ .*Chemical Relations.*

*Properties.*—Hippuric acid, known also as *uro-benzoic acid*, separates from hot solutions on cooling, in the form of minute spangles, or of larger, obliquely-striated, four-sided prisms, terminating at the ends in two flat surfaces (Fig. 11). The elementary form of the crystals is a vertical rhombic prism, which is best studied in microscopical crystals obtained by the slow evaporation of a solution of hippuric acid, which are similar

Fig. 11.



Hippuric acid from normal human urine, crystallized from water.

to those of phosphate of ammonia and magnesia, even in their most varied combinations (C. Schmidt).<sup>1</sup> This acid is devoid of smell, has a slightly bitter but not an acid taste, dissolves in 400 parts of cold water, and very freely in hot water; it is moreover readily soluble in alcohol, but difficult of solution in ether. Even the cold aqueous solution reddens litmus powerfully.

When gently heated, hippuric acid fuses, without loss of water, into an oily liquid, which, on cooling, solidifies into a crystalline milk-white mass; on the application of a stronger heat, there is produced a crystalline sublimate of benzoic acid and benzoate of ammonia, while a few oily drops are at the same time formed, which evolve an odor of cumarin (the oil of the Tonka bean), or fresh hay, solidify on cooling, and are soluble in alcohol and ammonia, but not in water. On exposing the acid to a more rapid and stronger heat, an intense odor of hydrocyanic acid is developed, and a porous coal is left as a residue.

Hippuric acid is unaffected by chlorine, chlorous, and dilute mineral acids; but when heated with concentrated hydrochloric or nitric acid, or even with oxalic acid, becomes decomposed (as already mentioned in page 142,) into benzoic acid and glycine (Dessaigne).<sup>2</sup> When heated

<sup>1</sup> Entwurf u. s. w. S. 36-40.<sup>2</sup> Compt. rend. T. 21, pp. 1224-1227.

with peroxide of manganese and sulphuric acid it is decomposed into carbonic acid, ammonia, and benzoic acid (Pelouze); boiled with freshly prepared peroxide of lead it yields benzamide, carbonic acid, and water (Fehling); and finally, if it be dissolved in nitric acid, and a stream of nitric oxide gas be passed through the solution, there is a development of ammonia, whilst there remains in solution a new non-nitrogenous acid which =  $C_{18}H_7O_7HO$  (Strecker).

Heated with hydrate of lime or caustic potash, hippuric acid yields benzine and ammonia, while the residue consists solely of carbonate of [lime or] potash, without a trace of cyanide of [calcium or] potassium. In fermenting and putrefying fluids this acid becomes decomposed into benzoic acid and other yet unknown products.

Shortly after Liebig's discovery of hippuric acid, while preparing it in large quantities from the urine of horses, I obtained one isolated crystal of hippuric acid half an inch in length, in which the vertical rhombic prism of the elementary form  $\infty P$  was combined with 2 micro-diagonal horizontal prisms, whereby the combining corners were truncated by the brachydiagonal horizontal prism. I have never again succeeded in obtaining crystals of such size and thickness.

*Composition.*—According to the above formula hippuric acid consists of:

Carbon, . . . . .	18 atoms,	. . . . .	60.335
Hydrogen, . . . . .	8 “ . . . . .	. . . . .	4.469
Nitrogen, . . . . .	1 “ . . . . .	. . . . .	7.821
Oxygen, . . . . .	6 “ . . . . .	. . . . .	22.347
Water, . . . . .	1 “ . . . . .	. . . . .	5.028
			<hr/> 100.000

The atomic weight of the hypothetical anhydrous acid = 2125.0; and its saturating capacity = 4.706.

From the various modes in which hippuric acid may be disintegrated, corresponding views have been taken of its constitution; all, however, agree in the opinion that in hippuric acid there must be concealed the radical *benzoyl*,  $C_6H_5$ , which is common to benzoic acid, volatile oil of bitter almonds, and benzamide. From the behavior of hippuric acid with peroxide of manganese and sulphuric acid, and from the composition of formobenzoic acid, which, as may be shown, consists of formic acid and oil of bitter almonds (hydride of benzoyl), Pelouze<sup>1</sup> concluded that hippuric acid was a kind of formobenzoic acid, which had assimilated hydrocyanic acid, so that it consisted of 1 equivalent of hydrocyanic acid, 1 equivalent of hydride of benzoyl, and 1 equivalent of formic acid, and =  $H.C_2N + H.C_6H_5 + C_2HO_3.HO$ .

This view of the composition of hippuric acid also finds some support in the circumstance that amygdalic acid, according to the recent investigations of Wöhler,<sup>2</sup> seems most probably to be formic acid, with oil of bitter almonds and sugar as an adjunct. If hippuric acid were actually composed in this manner, the products of decomposition with peroxide of manganese, could be hardly different from what they are, for hydro-

<sup>1</sup> Ann. de Chim. et de Pharm. T. 26, pp. 60-68.

<sup>2</sup> Ibid. Bd. 66, S. 238-242.

cyanic acid is very readily decomposed into formic acid and ammonia, and the oxygen yielded by the manganese converts the formic into carbonic acid, and the hydride of benzoyl into benzoic acid—both being processes of very frequent occurrence. But independently of the circumstance that, at least in analogous processes, some formic acid remains undecomposed, this view is also opposed by the fact that other oxidizing agents do not decompose hippuric acid in the same manner which they undoubtedly would do if the acid actually had this composition. On this account Fehling,<sup>1</sup> influenced by the behavior of hippuric acid with peroxide of lead, regarded it as fumaric acid conjugated with benzamide, and  $=\text{H}_2\text{N} \cdot \text{C}_{14}\text{H}_5\text{O}_2 + \text{C}_4\text{HO}_3 \cdot \text{HO}$ . If benzoic acid existed preformed in hippuric acid, it would be very unlikely that, by the action of an oxidizing agent, as peroxide of lead, a substance so poor in oxygen as benzamide should be formed.

Dessaigue's remarkable discovery must lead to the conclusion that glycine exists preformed in hippuric acid, and is conjugated with benzoic acid, so that 1 atom of anhydrous glycine with 1 atom of benzoic acid forms hydrated hippuric acid, since  $\text{C}_4\text{H}_4\text{NO}_3 + \text{C}_{14}\text{H}_5\text{O}_3 = \text{C}_{18}\text{H}_8\text{NO}_5 \cdot \text{HO}$ ; but if we are not altogether opposed to Strecker's formula for the formation of conjugated compounds from their constituents with the loss of certain atoms of water, yet it appears to us simple and natural that we should only compare with one another the formulæ of anhydrous combinations, and that certain atoms of water should not be arbitrarily abstracted; anhydrous glycine and anhydrous benzoic acid yield 1 atom of hydrogen, and 1 atom of oxygen more than anhydrous hippuric acid contains: if now, notwithstanding this, we assume that glycine exists preformed in hippuric acid, with however only a small quantity of water, we should proceed just as irrationally as if we assumed that ammonia existed in oxamide or in benzonitrile, because these bodies, when they assimilate water, yield ammonia. All, therefore, that we can maintain is, that in hippuric acid we find, in addition to benzoic acid, an adjunct  $=\text{C}_4\text{H}_3\text{NO}_2$ , which, on its separation, has a strong tendency to be transformed into glycine—a substance which is as readily formed as urea in the decomposition of nitrogenous matters (see pp. 142 and 147). It is in the changes which the adjunct undergoes in its intimate constitution by the action of stronger agents, that we must seek to ascertain the reason why the fixed acid is freed from the adjunct. This adjunct of hippuric acid might be regarded in reference to its composition, as an amide of fumaric acid ( $\text{C}_4\text{H}_3\text{NO}_2 = \text{H}_2\text{N} \cdot \text{C}_4\text{HO}_2$ ), and we should thus arrive at the reverse of Fehling's view of the subject. The question therefore now remains—Is it more probable that in hippuric acid benzamide is combined with fumaric acid, or fumaramide with benzoic acid? or is it more probable that in the action of peroxide of lead the benzoic acid is converted into benzamide by the oxidation of the fumaramide, or that by the action of concentrated acids the benzamide is decomposed and fumaramide formed? No satisfactory answer to these questions can be deduced either from the laws of stoichiometry or of affinity; since most unquestionable observations show in both cases the remarkable fact of the alternating substitution of 1 atom of amide and 1 atom of oxygen (for in the con-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 28, S. 48.



version of benzoic acid into benzamide the former takes in exchange 1 equivalent of amide for 1 atom of oxygen, and a similar substitution occurs in the conversion of fumaric acid into fumaramide). If, however, we regard benzoic acid as existing preformed in hippuric acid, we are by no means constrained to assume that the adjunct is fumaramide, or indeed any amide-compound. If we represent the formula of hippuric acid as  $C_4H_3NO_2 \cdot C_{14}H_5O_3 \cdot HO$ , this view is supported in the first place by the circumstance that hippuric acid has many physical and chemical properties in common with benzoic acid, which lead to the assumption that benzoic acid exists preformed in it, but afford no presumption in favor of the pre-existence of benzamide or fumaric acid in it. Secondly, we are indebted to the labors of Strecker for our knowledge of another conjugated acid, in whose analogous decomposition by acids glycine is also separated, which here also can only be produced by the assimilation of water; this acid being the biliary acid presently to be considered, where the same adjunct is combined with the cholic acid which we have already described. Thirdly, the fact discovered by Wöhler, that benzoic acid in its passage through the animal organism, is converted into hippuric acid, affords a certain amount of support to this view.

Recently, however, Strecker<sup>1</sup> has been led to yet another view regarding the constitution of hippuric acid, from its behavior with nitric oxide, and from the formation of the acid whose formula is  $C_{18}H_7O_7 \cdot HO$ . He looks upon hippuric acid as an amide-compound of this acid, and  $= H_2N \cdot C_{18}H_7O_7$ ; but the amides never have acid properties (besides which this only represents the hydrated hippuric acid); if Strecker had not ascertained that the silver-salt was accurately represented by  $AgO \cdot C_{18}H_7O_7$ , we might have regarded its composition as expressed by the formula  $C_9H_3O_3 \cdot HO$ , and therefore have considered hippuric acid as analogous to oxamic, lactamic, tartramic, and aspartic acids, and as a compound of this acid with its amide ( $H_2N \cdot C_9H_3O_2 + C_9H_3O_3 \cdot HO = C_{18}H_8NO_5 \cdot HO$ ). The view, in accordance with which benzoic acid exists preformed, is, however, still the most probable.

*Combinations.*—With alkalis and alkaline earths hippuric acid forms crystallizable salts soluble in water and having a bitter taste; its combinations with metallic oxides are difficult of solution in cold water, but dissolve somewhat more freely in hot water. All the crystallized salts contain water of crystallization. Schwartz<sup>2</sup> has analyzed the following salts:

*Neutral hippurate of potash*,  $KO \cdot \bar{H}i + 2HO$ , occurs in microscopic, oblique rhombic prisms, which part with their water at  $100^\circ$ . The acid salt  $KO \cdot \bar{H}i + HO \cdot \bar{H}i + 2HO$ , crystallizes in broad, satiny plates.

*Hippurate of soda*,  $2NaO \cdot \bar{H}i + HO$ , is crystalline, and dissolves readily in water and alcohol.

*Acid hippurate of ammonia*,  $H_4NO \cdot \bar{H}i + HO \cdot \bar{H}i + 2HO$ , occurs in very minute, four-sided, square prisms; it behaves, when thrown upon water, like butyrate of baryta.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 58.

<sup>2</sup> Ibid. Bd. 54, S. 29–51. [Schwartz has published another memoir on this acid during the last few months (in Ann. d. Ch. u. Pharm. Bd. 75, S. 190).—a. r. d.]

*Hippurate of baryta*,  $\text{BaO} \cdot \text{Hi} + \text{HO}$ , is obtained in microscopic, square prisms, and loses its water at  $100^\circ$ .

*Hippurate of strontia*,  $\text{SrO} \cdot \text{Hi} + 5\text{HO}$ , occurs in broad plates, difficult of solution in cold water, or in microscopic, four-sided prisms, with large terminal planes.

*Hippurate of lime*,  $\text{CaO} \cdot \text{Hi} + 3\text{HO}$ , occurs in oblique rhombic prisms; it parts with all its water at  $100^\circ$ .

*Hippurate of magnesia*,  $\text{MgO} \cdot \text{Hi} + 5\text{HO}$ , crystallizes in wart-like masses, is readily soluble, and at  $100^\circ$  loses only 4 atoms of water.

*Hippurate of cobalt*,  $\text{CoO} \cdot \text{Hi} + 5\text{HO}$ , occurs in rose-colored wart-like masses, consisting of microscopical, flat, four-sided prisms; at  $100^\circ$  it loses all its water; and it is perfectly insoluble in alcohol.

*Hippurate of nickel*,  $\text{NiO} \cdot \text{Hi} + 5\text{HO}$ , forms apple-green crusts, dissolves in warm spirit, and at  $100^\circ$  loses all its water.

*Hippurate of copper*,  $\text{CaO} \cdot \text{Hi} + 3\text{HO}$ , occurs in blue, oblique rhombic prisms, and at  $100^\circ$  is anhydrous.

*Hippurate of lead*,  $\text{PbO} \cdot \text{Hi}$ , crystallizes from hot solutions with 2 atoms of water in fine silky tufts of needles; from cold solutions, by slow evaporation, in broad four-sided tablets, with 3 atoms of water. At  $100^\circ$  it is anhydrous.

*Hippurate of silver*,  $\text{AgO} \cdot \text{Hi} + \text{HO}$ , occurs as a curdy precipitate, which dissolves in boiling water, and, on cooling, separates in beautiful silky needles.

*Hippurate of iron* occurs as a dingy, voluminous precipitate, which does not dissolve, but fuses in boiling water; it dissolves in warm alcohol, but falls as an amorphous precipitate on cooling; it crystallizes from the cold solution in oblique rhombic prisms.

*Hippurate of oxide of ethyl*,  $\text{C}_4\text{H}_5\text{O} \cdot \text{C}_{18}\text{H}_8\text{NO}_5$ , forms long, white, silky needles, with a greasy feeling, devoid of odor, of an acrid taste, slightly soluble in cold, but more so in hot water; it fuses at  $44^\circ$ , solidifying again at  $32^\circ$ , and on exposure to a stronger heat it decomposes.

*Products of its metamorphosis*.—The non-nitrogenous acid,  $\text{C}_{18}\text{H}_7\text{O}_7 \cdot \text{HO}$ , obtained from hippuric acid by the action of nitrous acid, is, according to Strecker, readily soluble in ether, yields with baryta a salt, crystallizing in silky needles, and readily soluble in water, and with oxide of silver a salt,  $\text{AgO} \cdot \text{C}_{18}\text{H}_7\text{O}_7$ , which dissolves in boiling water, and on cooling crystallizes in delicate needles; and which, on exposure to heat, develops hydride of benzoyl. The production of this acid from hippuric acid is shown in the equation  $\text{C}_{18}\text{H}_8\text{NO}_5 + 3\text{HO} - \text{H}_3\text{N} = \text{C}_{18}\text{H}_7\text{O}_7 \cdot \text{HO}$ .

*Preparation*.—Hippuric acid is very easily obtained from the urine of horses, but there is some difficulty in separating it from the coloring matter. Fresh urine, obtained from horses, is evaporated to  $\frac{1}{3}$ th of its volume, and then treated with hydrochloric acid; after it has cooled, the acid which has separated, and is usually much discolored, is dissolved in ten times its bulk of boiling water, and boiled with milk of lime; the solution is filtered, a solution of alum is added till there is an acid reaction, and the alumina is then precipitated by bicarbonate of soda. The boiling with milk of lime destroys a portion of the pigment adhering to

the hippuric acid, while another portion of the pigment is precipitated with the alumina. The acid precipitated by hydrochloric acid from the filtered fluid is again dissolved in boiling water, boiled with animal charcoal, and filtered while hot; on cooling, the acid now separates in a colorless state. Moreover, by mere, but often repeated boiling of horses' urine, and of the hippuric acid separated from it with milk of lime, we may obtain it free from color.

Perfectly fresh urine must be used, since horses' urine, even at an ordinary temperature, very soon begins to decompose; and it then no longer yields hippuric but benzoic acid.

*Tests.*—Hippuric acid presents such characteristic properties, that if it be once pretty well freed from other substances, it can scarcely be confounded with any other acid, except, perhaps, benzoic acid, if the latter be contaminated with organic coloring, and nitrogenous matters; since in the pure state the two acids act so differently when exposed to heat that it is impossible to confound one with the other.

When they occur in an impure state, they may be distinguished from one another by attention to the following points.

Hippuric acid, which is far less soluble in ether than benzoic acid, crystallizes from hot saturated solutions in needles or prisms, while benzoic acid crystallizes in scales. The latter often causes such a solidification of the whole fluid, that the vessel after cooling may be inverted without the escape of a single drop. Further, on the addition of acids to solutions of their salts, hippuric acid is at once precipitated in needles or spangles, while benzoic acid gives rise to a milky turbidity before it crystallizes. On rapidly evaporating an acid fluid in a basin covered over with paper, delicate glistening scales may be observed on its lower surface if benzoic acid be present, but not if hippuric acid alone be present in the fluid. The microscope, however, affords the best means of distinguishing these acids from one another, by comparing their crystalline forms in accordance with the directions given in pp. 83 and 173. With such an examination, it is impossible that these acids can be confounded.

In order to detect small quantities of hippuric acid in animal fluids, we must be especially careful that such fluids are fresh, since if this be not the case, the hippuric acid will have become changed into benzoic acid, which on evaporation for the most part escapes with the aqueous vapor; if, however, the animal fluid be still perfectly undecomposed, it must be evaporated to almost the consistence of a syrup and then extracted with alcohol of specific gravity, 0.83; a little oxalic acid must be added to the alcoholic extract during its evaporation, which must be continued till it assumes a syrupy consistence; the residue must then be extracted with ether to which  $\frac{1}{6}$ th of its volume of alcohol has been added. This extract must now be carefully evaporated, and the residue, which, besides free acids, also contains fatty matters, must be treated with water in order to remove the latter. It sometimes happens that on the addition of the water, crystals of hippuric acid at once separate from the above extract-like mass; but whether this be the case or not, this ethereal extract must be warmed with water, and allowed to percolate through a previously well-moistened filter; the filtered acid fluid may

then either be gently concentrated by warmth, or if its quantity be very small, it may be left to spontaneous evaporation in a watch-glass; crystals of hippuric acid very soon separate, whose form must be determined by the microscope. If much hippuric acid be present, it will sometimes separate from the syrupy residue by the mere addition of hydrochloric acid, and can be distinguished from uric acid and other crystalline substances by the microscope.

### *Physiological Relations.*

*Occurrence.*—Hippuric acid was first recognized by Liebig as an independent acid in *horses' urine* where it had previously been mistaken for benzoic acid; it has been subsequently found in the urine of many graminivorous animals, as, for instance, oxen, elephants, goats, hares, sheep, &c. It is, however, singular that, according to Wöhler, it is entirely absent in the urine of calves while suckling, although the fluid contains allantoin, uric acid, and urea (see p. 163). In the urine of the pig neither Boussingault,<sup>1</sup> nor von Bibra,<sup>2</sup> could discover any hippuric acid. Liebig<sup>3</sup> was the first who recognized its presence in healthy human urine, in which it principally occurs after the use of vegetable food: according to him it exists in human urine, in about the same quantity as uric acid, while according to Bird,<sup>4</sup> the hippuric acid most commonly stands to the uric acid in the ratio of 1 : 3.

I have already remarked in p. 83, that benzoic acid never occurs in fresh horses' urine, and that it is merely a product of the decomposition of that fluid; I can, however, perfectly confirm the observation of Schmidt,<sup>5</sup> that hippuric acid is occasionally, although very rarely, entirely absent, and that in its place there is found an oily matter which when heated with caustic alkalies yields benzene.

Attempts have been made to refute Liebig's assertion that hippuric acid always exists in human urine, at least after the use of vegetable food; but although I formerly did not succeed in detecting this acid in my own urine during a purely vegetable diet, I have since very frequently convinced myself, from experiments both on large and on small quantities of urine, that this acid is constantly present during the use of a mixed diet. The presence of hippuric acid may, however, readily escape our notice if we evaporate the acid urine too rapidly, after the acid has been converted into benzoic acid; on the other hand, we need be under no apprehension that the hydrochloric acid which is added will decompose the hippuric acid, as in order to effect any change on it, a very concentrated acid and prolonged boiling are required.

Hippuric acid is not found in the urine of *carnivorous animals*, but it has probably not been sought for with sufficient care and attention. In the urine of tortoises neither J. Müller and Magnus,<sup>6</sup> nor Marchand<sup>7</sup>

<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. T. 15, pp. 97–104.

<sup>2</sup> Ann. de Ch. u. Pharm. Bd. 53, S. 98–112.

<sup>3</sup> Ibid. Bd. 37, S. 257.

<sup>4</sup> London Medical Gazette, vol. 34, p. 685: [In his Urinary Deposits, &c., 3d edit. p. 96, this opinion is considerably modified. We there find that "its quantity in health is not constant, and always, unless after the ingestion of benzoic or cinnamic acid, very much less than has been stated."]—G. E. D.]

<sup>5</sup> Entwurf u. s. w. S. 39.

<sup>6</sup> Müller's Archiv. 1835. S. 214.

<sup>7</sup> Journ. f. pr. Ch. Bd. 34, S. 244–247.

could detect hippuric acid ; I have, however, convinced myself with the greatest certainty, and on many occasions, that hippuric acid is present in addition to uric acid in the urine of *Testudo græca*.

Magnus was unable even to find uric acid in the urine of *Testudo nigra* s. *elephantopus*, while Marchand found uric, but no hippuric acid in the urine of *T. tabulata* ; I probably worked with much larger quantities, and certainly always used fresh urine. My specimens of *Testudo græca* were fed with lettuce and other vegetables. The urine may be easily collected by placing the animal on its back in a dish ; when the bladder is moderately filled, the animal very soon spontaneously\* passes its urine, which besides alkaline urates and hippurates, contains free hippuric acid. Without the preliminary addition of a stronger acid, we may obtain the hippuric acid in a crystalline state by the addition of water to the ethereal extract, and sufficiently pure to admit of our accurately studying its behavior when exposed to heat, its solubility, &c. ; if, however, oxalic or hydrochloric acid were used in the process, in the manner which has been already explained, we should obtain much larger quantities of hippuric acid.

It has been already mentioned (p. 84) that both Wöhler and Ure discovered that benzoic acid was converted in the animal organism into hippuric acid, and eliminated as such with the urine. The subsequent observations of Erdmann and Marchand<sup>1</sup> having shown that cinamic acid undergoes a similar metamorphosis, it became a point of interest to ascertain whether the other acids homologous to benzoic acid, namely, toluylic and cumic (or cuminic) acids, were also transformed into hippuric acid ; but this is by no means the case, as is shown by the independent investigations of Hoffmann<sup>2</sup> and of Ranke.<sup>3</sup> Moreover, the acids which are homologous to it in their amount of carbon and hydrogen, as salicylic acid, anisic acid, and cumaric acid, pass, like those previously mentioned, in an unchanged state into the urine, as was shown by Ranke's experiments.

In *morbid human urine* I have almost always been able to detect hippuric acid ; it especially occurs in large quantity in acid *febrile urine*, whether the fever be typhus or be associated with pneumonia or any other pathological process. Before hippuric acid was discovered in healthy human urine, I detected its presence in *diabetic urine*,<sup>4</sup> in which it is more easily recognized than in other forms of urine which abound in extractive matters.

In *diabetic urine* I have found hippuric acid in every instance in which I have sought for it ; Ambrosiani, Hünefeld, and others have also found it in the urine during this disease ; Bouchardat found it in a case of what is called *diabetes insipidus* ; Pettenkofer<sup>5</sup> found it in large quantity in the urine of a girl with chorea. In the case of a drunkard with a contracted, probably a hob-nail, liver, Bird<sup>6</sup> observed a sediment consisting of hippuric acid, on the addition of hydrochloric acid to the concentrated urine. In the strongly acid urine which is sometimes

<sup>1</sup> Journ. f. pr. Ch. Bd. 35, S. 307-309.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 74, S. 342.

<sup>3</sup> Journ. f. pr. Ch. Bd. 56, S. 3-6.

<sup>4</sup> Journ. f. pr. Ch. Bd. 6, S. 113.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 128.

<sup>6</sup> London Medical Gazette, vol. 34, p. 686.

passed in fevers, the acid reaction is in a great degree dependent on the hippuric acid; from the ethereal extract of urine of this nature, and without the preliminary addition of any acid, we often obtain the most beautiful crystals of hippuric acid. Such urine is, however, by no means so common as is generally supposed; for this febrile urine is much more rapidly rendered acid by lactic acid (which is not formed till after the emission of the urine), than the normal secretion, and hence, unless it be examined when perfectly fresh, we usually find that febrile urine is more acid than the normal fluid. I have not been able to establish any relation between certain morbid processes or groups of symptoms and the amount of the hippuric acid contained in the urine.

Hippuric acid has as yet been found nowhere but in the urine. [Its recent discovery in the blood of oxen, by Verdeil and Dollfuss, is noticed in the second volume, in the article on "The Blood."—G. E. D.]

*Origin.*—Notwithstanding the many points which seem to elucidate the inquiry, the formation of hippuric acid in the animal body still remains unexplained. All views regarding the chemical constitution of hippuric acid coincide in the belief that it contains, hidden within it, a benzoyl-compound ( $C_{14}H_5O_2 + II$  or  $+O$  or  $+H_2N$ ); it is an established fact that benzoic acid, oil of bitter almonds, and cinnamic acid, which is very similar to benzoic acid, are transformed in the animal body into hippuric acid. Now, since the benzoyl-compounds are almost entirely confined to the vegetable kingdom, we might believe that this constituent of hippuric acid principally arises from vegetable food, and the abundance of this acid in the urine of many herbivorous animals is in favor of this view. We might therefore be led to regard one constituent of hippuric acid as an immediate product of decomposition of certain constituents of food, namely, of the vegetable portion; but this view is opposed by several positive experimental results; thus in the urine of patients on an antiphlogistic diet, who for several days have scarcely taken any food, the amount of hippuric acid is actually increased.

The urine of tortoises, which had been kept fasting for more than six weeks, still contained hippuric acid; and it occurred in the urine of diabetic patients who were restricted to a purely animal diet. In the urine of granivorous birds, as well as in that of the larva of *Sphinx Cossus*, and of several other herbivorous insects, I have found, after careful examination, larger or smaller quantities of uric acid, but no hippuric acid. Hence we may conclude in the first place that the formation of uric acid is not dependent on the use of animal food, or that of hippuric acid on the use of vegetable food, and secondly, that the latter acid must derive its nitrogenous constituent from the retrograde metamorphosis of the animal tissues. This is, moreover, not opposed to our chemical facts in relation to the production of the benzoyl-compounds, for there is every reason to believe that the nitrogenous tissues which, according to the admirable investigations of Guckelberger, when treated with oxidizing agents, yield benzoic acid and benzonitrile, yield a like product of decomposition during the gradual oxidation which they undergo in the animal body.

<sup>1</sup> [Compt. rend. T. 29, p. 789; and more fully in the Ann. d. Ch. u. Pharm. Bd. 74, S. 214.]

In reference to the nitrogenous constituent of hippuric acid we may regard it as fumaramide, or as glycine; it is undoubtedly derived from the animal albuminous substances, and probably from effete tissue. It would, however, certainly be rash to attribute it principally to the decomposition of the gelatigenous tissues, simply because it is chiefly formed from them in artificial experiments; but independently of the circumstance that this product into which the nitrogenous adjunct of hippuric acid becomes converted, may also be obtained from albuminous substances, we must bear in mind that the metamorphosis going on in the gelatigenous tissues is certainly too insignificant to account for the quantity of hippuric acid found in the urine (as, for instance, after the ingestion of from two drachms to half an ounce of benzoic acid), and that the same substance is separated even more abundantly from the liver. Glycine must therefore be regarded in the same light as urea, as a common product of decomposition of nitrogenous substances.

We cannot therefore find any very immediate source from which either of the proximate constituents of this acid can be derived, since neither physiological nor pathological relations elucidate the process by which it is formed in the animal body.

This much, however, is certain, that hippuric acid is to be regarded merely as a product of excretion, and consequently that it can have no special uses in the animal organism. \*

It is to be regretted that benzoic acid is so rarely prescribed by the physician; and that, even in those cases, it is usually ordered on most irrational principles. It deserves to be thoroughly tested in a pharmacological point of view; it certainly possesses one great advantage over all the other officinal acids in its property of rendering the urine strongly acid. Ure attaches great importance to this circumstance, but it does not appear to have been turned to much account in actual practice.

## URIC ACID.— $C_5H_4N_2O_2.HO$ .

### *Chemical Relations.*

*Properties.*—Pure uric acid occurs either in a glistening white powder, or in very minute scales, which under the microscope are seen to consist of irregular plates, whose crystalline form (see our remarks on the crystals, in the consideration of the "Tests,") cannot very well be made out: it is a substance devoid of odor and taste; it requires 1800 or 1900 parts of hot, and 14,000 or 15,000 parts of water at the ordinary temperature of  $20^\circ$ , to dissolve it; it is insoluble in alcohol and ether, and does not redden litmus. It dissolves in concentrated hydrochloric acid somewhat more readily than in water; it dissolves tolerably freely, and without decomposition, in concentrated sulphuric acid, but is again precipitated on the addition of water. It dissolves readily in the alkaline carbonates, borates, phosphates, lactates, and acetates, since it abstracts some of the alkali from these salts, and is thus rendered more soluble. Uric acid is expelled from all its salts by acetic as well as by other acids, and on its separation at first forms a gelatinous mass (according to

Fritzsche,<sup>1</sup> a hydrate =  $C_5H_4N_2O_2 \cdot HO + 4HO$ ), which, however, soon changes into small glistening plates.

Uric acid belongs to the *weakest class of acids*; thus, as in the case of the fatty acids, it does not directly expel carbonic acid from carbonate of potash, but urate of potash and bicarbonate of potash are formed, if a sufficient amount of uric acid be added; if the solution of potash be concentrated, the urate of potash remains undissolved; the behavior of uric acid to the alkaline borates and phosphates is similar, with the exception of this difference, that the solution of phosphate of soda, which has an alkaline reaction, reddens litmus when an excess of uric acid has been added to it, in consequence of the formation of biphosphate of soda.

Uric acid, when submitted to *dry distillation*, is converted into urea, cyanic acid, cyamelide, hydrocyanic acid, and a little carbonate of ammonia, leaving, as a residue, a brownish-black coal, rich in nitrogen.

On fusing uric acid with *hydrated potash*, carbonate and cyanate of potash, with cyanide of potassium, are formed. On boiling uric acid with 20 parts of water, and adding peroxide of lead as long as the brown color of the oxide continues to disappear, there are formed oxalate of lead, urea, and allantoine ( $2C_5H_4N_2O_2 \cdot HO + 2O + 3HO = C_2H_4N_2O_2 + 2C_2O_3 + C_4H_3N_2O_3$ ).

Moist uric acid, placed in *chlorine gas*, intumescs, and, giving off carbonic and cyanic acids, is converted into oxalic acid and hydrochlorate of ammonia; dry uric acid in dry chlorine gas yields much cyanic acid, chloride of cyanogen, and hydrochloric acid, leaving only a small carbonaceous residue. Uric acid dissolves with considerable readiness in dilute *nitric acid*, developing equal volumes of nitrogen and carbonic acid, and yielding to the solution several of the different products of decomposition which we shall presently describe. On evaporating to dryness a solution of uric acid in nitric acid, there is left a red amorphous residue, which, especially if we expose it to the vapor of ammonia, assumes a very beautiful purple tint; on moistening the red mass (murexide) with a little caustic potash, a beautiful violet tint is developed (Schlossberger).<sup>2</sup>

*Composition*.—According to the above formula, deduced by Bensch<sup>3</sup> from his analyses of the urates, uric acid consists of:

Carbon,	.	.	.	.	5 atoms,	.	.	.	.	35.714
Hydrogen,	.	.	.	.	1 "	.	.	.	.	1.191
Nitrogen,	.	.	.	.	2 "	.	.	.	.	33.833
Oxygen,	.	.	.	.	2 "	.	.	.	.	19.048
Water,	.	.	.	.	1 "	.	.	.	.	10.714
										<hr/>
										100.000

The atomic weight of the hypothetical anhydrous acid = 937.5, and its saturating capacity = 10.656. There is hardly any other organic acid, whose products of decomposition have been so accurately and so generally examined as those of uric acid, and yet chemists have been unable to establish for it any rational formula. Bensch's discovery of

<sup>1</sup> Bull. scient. de St. Petersb. T. 1, Nos. 79 et 107.

<sup>2</sup> Arch. f. physiol. Heilk. Bd. 8, S. 294.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 51, S. 189-208.



the true atomic weight of uric acid has tended to weaken the views which were previously held regarding the intimate constitution of this acid. If we choose to double the atoms, and if we so far extend the idea of conjugation, that the conjugating substances may, in their union, lose certain atoms of hydrogen and oxygen (so that we might regard oxamide as a body composed of oxalic acid and ammonia, and benzanilide as composed of benzoic acid and aniline), then indeed, much might be explained at which we could not arrive by a strict logical induction. Taking into consideration those substances which for a long time have been regarded as conjugated, it seems that we should only consider as true conjugated bodies those compounds in which two organic bodies unite with one another, the union being accompanied with a loss of water; which, however, in some cases may be shown by direct experiment, and in others, may be assumed with great probability, to lie without the true atomic group, and may therefore be regarded as a basic acid, or saline atom of water. Many of the substances which have been recently regarded as conjugated bodies, undoubtedly contain certain atoms of oxygen and hydrogen less than the anhydrous substances from which they are produced, or may be supposed to be produced; but this view does not coincide with the original idea of a conjugated body; especially when it is probable that in this union one of the substances has contributed the oxygen and the other the hydrogen for the formation and separation of the water.

It would be equally injudicious, were we not to facilitate the recognition of the metamorphosis or transposition of the atoms of organic substances, by some general remarks on the connection and separation of atoms.

Such remarks, however, are not based on anything more than a fiction, and do not rest on a conclusion obtained by induction. That such hypotheses are not always to be rejected in the natural sciences, is shown by Newton's hypothesis of emanating rays of light, which now, indeed, is entirely displaced by the undulatory theory. In this light we must consider the view regarding the composition of uric acid, put forth some years ago by Liebig and Wöhler. From the decomposition of uric acid by peroxide of lead, they deduced, for uric acid, the hypothetical formula,  $C_2H_4N_2O_2 + 2C_4NO_2$ ; that is to say, they regarded urea as existing preformed in it, together with an acid incapable of isolation in an undecomposed state, to which they applied the name of *urilic acid*. Now that the substratum of this hypothesis has been more than shaken by the discovery of the true atomic weight of uric acid, we may yet make use of this fiction in order to be able to represent the formation of the products enumerated by Liebig and Wöhler in their classical investigations regarding uric acid. Thus we may conceive, that on the decomposition by peroxide of lead, 2 equivalents of hydrated uric acid contain 1 equivalent of urea, which is isolated, while the 2 equivalents of urilic acid are, in the first place, decomposed into  $C_4O_4$  and  $C_4N_2$ , of which the former assimilates 2 atoms of oxygen, and forms oxalic acid, while the latter assimilates 3 atoms of water, and produces allantoin. In a similar way we can elucidate the mode of formation of those numerous products which result from the action of nitric acid on uric acid.

*Combinations.*—It is only with the fixed alkalis that uric acid forms

salts which possess even a moderate degree of solubility; the lithia-salt is especially soluble, while urate of ammonia is almost insoluble. Potash and soda are the only bases with which uric acid forms neutral salts; with ammonia and all other bases it forms only acid and insoluble salts. On passing carbonic acid into a potash-solution of uric acid, an acid potash-salt is precipitated.

*Neutral urate of potash*,  $\text{KO.C}_5\text{HN}_2\text{O}_2$ , is obtained on mixing alcohol with a solution of uric acid in potash (free from the carbonate) and concentrating the solution. It crystallizes in needles free from water, dissolves in 30 or 40 parts of boiling water, slightly in alcohol, and not at all in ether, has a strong alkaline reaction, and attracts carbonic acid from the atmosphere.

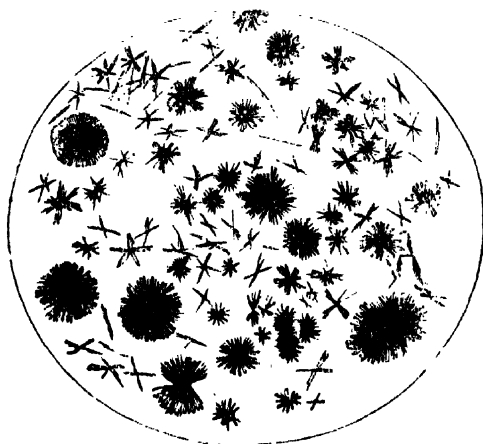
*Bi-urate of potash*,  $\text{KO.C}_5\text{HN}_2\text{O}_2 + \text{HO.C}_5\text{HN}_2\text{O}_2$ , is precipitated by carbonic acid from the solution of the neutral salt; it crystallizes in needles, dissolves in 70 or 80 parts of boiling water, and in 700 or 800 parts of water at  $20^\circ$ . The solution does not exhibit an alkaline reaction, and is precipitated by hydrochlorate of ammonia and the alkaline bicarbonates.

*Neutral urate of soda*,  $\text{NaO.C}_5\text{HN}_2\text{O}_2 + \text{H}_2\text{O}$ , crystallizes in wart-like masses, dissolves in 80 or 90 parts of boiling water, is slightly soluble in alcohol, and insoluble in ether; at  $100^\circ$  it loses its water of crystallization.

*Bi-urate of soda*,  $\text{NaO.C}_5\text{HN}_2\text{O}_2 + \text{HO.C}_5\text{HN}_2\text{O}_2 + \text{H}_2\text{O}$ , crystallizes in short hexagonal prisms, or in thick six-sided (microscopic) tablets, which commonly arrange themselves in star-formed masses in which the individual crystals are larger and can be more distinctly made out than in the microscopic aggregations of the ammonia-salt; it begins to lose its water of crystallization at  $170^\circ$ , and is soluble in 124 parts of boiling and 1150 parts of cold water.

*Bi-urate of ammonia*,  $\text{H}_4\text{NO.C}_5\text{HN}_2\text{O}_2 + \text{HO.C}_5\text{HN}_2\text{O}_2$ , may be ob-

Fig. 12.



Bi-urate of Ammonia.

tained crystallized in extremely delicate needles, but it also forms under the microscope, globular opaque masses, from some points of which extremely delicate spikelets may be seen to project.

Almost all the other salts of uric acid occur as amorphous precipitates, and consist of 1 atom of base and 2 atoms of uric acid, of which 1 atom always retains its basic atom of water; hence we cannot well assume that the atomic weight of uric acid should be doubled (that is to say  $=C_{10}H_2N_4O_4$ ), for if, with such an atomic weight, these salts were all neutral salts, they, or at all events, some of them, would certainly lose this 1 atom of water at a higher temperature.

The salts of *baryta*, *strontia*, and *lime*, are represented by the formula  $RO.C_5HN_2O_2 + HO.C_5HN_2O_2 + HO$ .

*Bi-urate of magnesia*,  $MgO.C_5HN_2O_2 + HO.C_5HN_2O_2 + 6HO$ , crystallizes in delicate needles, loses 5 of its 6 atoms of water at  $170^\circ$ , and dissolves in 160 parts of boiling water, but in not less than 3800 parts of cold water.

*Bi-urate of lead*,  $PbO.C_5HN_2O_2 + HO.C_5HN_2O_2 + HO$ , is a white powder, which loses its water of crystallization at  $160^\circ$ .

*Bi-urate of copper*,  $CaO.C_5HN_2O_2 + HO.C_5HN_2O_2 + 5HO$ , is a green powder which, at  $140^\circ$ , loses 3 atoms of water of crystallization.

*Sulphate of uric acid*,  $HO.C_5HN_2O_2 + 4 (HO.SO_3)$ , is formed by dissolving uric acid in warm concentrated sulphuric acid, from which, on cooling, it separates in colorless crystals, which fuse at  $70^\circ$ , in cooling again solidify in a crystalline mass, and become decomposed at  $170^\circ$ ; it attracts water from the atmosphere, and thus becomes decomposed into its proximate constituents in the same manner as if water were added to it.

*Products of its metamorphosis.*—The products of decomposition of uric acid are of extreme interest, insomuch as they afford us a deep and general insight into the various transpositions of atoms and atomic groups.

*Alloxan, erythric acid*,  $C_8H_4N_2O_{10}$ , is produced when 1 part of dry uric acid is gradually introduced into 4 parts of nitric acid of 1.42 to 1.53 specific gravity, when the whole finally solidifies and becomes crystalline. A better method of preparing this body is by mixing 4 parts of uric acid with 8 parts of moderately strong hydrochloric acid, and then gradually introducing 1 part of chlorate of potash into the fluid; in the latter case, urea and alloxan are formed without any development of gas, while in the former case, nitrogen and carbonic acid are evolved in consequence of the decomposition of the urea by nitrous acid. (Compare p. 144.)

Alloxan crystallizes in large colorless rhombic octohedra (which at first have a diamond-like lustre, but soon effloresce) with 6 atoms of water of crystallization from hot but not perfectly saturated solutions; while from saturated solutions it crystallizes in anhydrous four-sided prisms: it has a faintly saline taste, a sickly odor, reddens litmus, and communicates a purple-red color to the skin.

It is easy to see that in accordance with the above fiction respecting urilate of urea, urilic acid assimilates 4 atoms of water and 2 atoms of oxygen, and thus forms alloxan ( $C_8N_2O_4 + 4HO + 2O = C_8H_4N_2O_{10}$ ).

*Alloxanic acid*,  $C_8HNO_4.HO$ , is formed by digesting alloxan with caustic alkalies, and by decomposing the baryta-salt by sulphuric acid. It crystallizes in concentrically grouped needles, which are unaffected by

exposure to the atmosphere, have an acid (but subsequently leave a sweetish) taste, dissolve readily in water, less in alcohol, and very slightly in ether; this acid reddens litmus strongly, decomposes carbonates and acetates, and oxidizes zinc and cadmium, hydrogen being at the same time developed; in an aqueous solution it becomes decomposed at a temperature above  $60^{\circ}$ . Its alkaline salts are soluble in water and crystallizable; its other neutral salts are difficult of solution: like uric acid it has a strong tendency to form acid salts, all of which are soluble (Schlieper).<sup>1</sup>

Alloxanic acid is produced by the abstraction of 2 atoms of water from alloxan.

If a solution of alloxanic acid be submitted to prolonged ebullition, it evolves carbonic acid, and is decomposed into an acid insoluble in water, *leucoturic acid*,  $C_6H_3N_2O_6$ , and into a soluble indifferent body, *diffluan*;  $C_6H_4N_2O_5$  (Schlieper).

Two atoms of alloxan yield 1 atom of this new acid, and 1 atom of diffluan, besides 4 atoms of carbonic acid and 1 atom of water, for  $C_{16}H_8N_4O_{20} = C_6H_3N_2O_6 + C_6H_4N_2O_5 + 4CO_2 + HO$ .

*Mesoxalic acid*,  $C_3O_4$ , is produced together with urea, when a solution of alloxan is added by drops to a boiling solution of acetate of lead: it is crystallizable, and reddens litmus.

Alloxan becomes simply decomposed into 1 equivalent of urea and 2 equivalents of mesoxalic acid, for  $C_2H_4N_2O_2 + 2C_3O_4 = C_8H_2N_2O_{10}$ .

*Mykomelinic acid*,  $C_8H_5N_4O_5$ , is formed when an excess of dilute nitric acid is added to a supersaturated solution of alloxan, and boiled for some time with ammonia; in its moist state it occurs as a yellow gelatinous mass; when dried, it is a yellow powder, which is soluble in water, reddens litmus, and decomposes carbonates.

This acid is formed from 1 atom of alloxan and 2 atoms of ammonia with the separation of 5 atoms of water;  $C_8H_4N_2O_{10} + 2H_3N - 5HO = C_8H_5N_4O_5$ .

*Parabanic acid*,  $C_6N_2O_4 + 2HIO$ , is prepared by digesting 1 part of uric acid or of alloxan, with 8 parts of moderately diluted nitric acid, and evaporating the solution to the consistence of a syrup; after some time there is a separation of small plates or minute prisms of parabanic acid; it is unaffected by exposure to the atmosphere, has an acrid, sour taste, dissolves readily in water, fuses when heated, and partially sublimes without decomposition.

Parabanic acid is produced in the following manner from uric acid and nitric acid: the urea of the uric acid is decomposed as usual by the nitrous acid which is formed, but 2 atoms of water and 4 atoms of oxygen enter into combination with the urilic acid with which they form 2 atoms of carbonic acid, and 1 atom of parabanic acid, for  $C_8N_2O_4 + H_2O_2 + 4O - C_2O_4 = C_6N_2O_4 + 2HO$ .

*Alloxan* with 2 atoms of oxygen becomes decomposed into 2 atoms of carbonic acid, 4 atoms of water, and 1 atom of parabanic acid, for  $C_8H_4N_2O_{10} + 2O = C_2O_4 + H_4O_4 + C_6N_2O_4$ .

*Hydrurilic acid*,  $C_{12}H_3N_3O_9 + 2HO$ , is formed at the same time with

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 55, S. 251-297.

alloxan under certain conditions not yet accurately understood; it occurs as a white flocculent powder, consisting of delicate needles; it is difficult of solution in cold, but dissolves more readily in hot water; it is insoluble in alcohol; with the alkalis it forms acid and neutral salts; this acid may be regarded as a combination of the above-mentioned hypothetical urilic acid with water; 3 atoms of urilic acid and 10 atoms of water forming 2 atoms of hydrurilic acid. By nitric acid, this acid is converted into nitro-hydrurilic acid,  $C_8H_2N_3O_{14}$ .

*Oxaluric acid*,  $C_6H_3N_2O_7.HO$ : if a solution of uric acid in dilute nitric acid be supersaturated with ammonia and evaporated, the ammonia-salt of this acid separates in needles; on separating the acid from the salt by means of a more powerful acid we obtain it as a glistening white crystalline powder with an acid taste and an acid reaction; when heated it becomes decomposed into 2 atoms of oxalic acid and 1 atom of urea, for  $C_6H_3N_2O_7.HO = 2C_2O_3 + C_2H_4N_2O_2$ .

*Crystallized oxaluric acid* may therefore be regarded as a combination of 2 atoms of oxalic acid and 1 atom of urea, for  $C_4O_6 + C_2H_4N_2O_2 = C_6H_4N_2O_8$ .

Parabanic acid when boiled with ammonia takes up 3 atoms of water, and forms *oxaluric acid*, for  $C_6N_2O_4 + 11_3O_3 = C_6H_3N_2O_7$ .

*Thionuric acid*,  $C_8H_7N_3S_2O_{14}$ , is formed by mixing a solution of alloxan with an excess of aqueous sulphurous acid, supersaturating with ammonia and boiling for some time; as the solution cools, *thionurate of ammonia* separates in nacreous crystalline scales; on combining the acid of this salt with lead, decomposing the lead-salt by sulphuretted hydrogen, and evaporating the filtered fluid, we obtain *thionuric acid* in the form of a white crystalline mass with an acid taste, which is unaffected by exposure to the air, dissolves readily in water, and is decomposed both by simple boiling and on the addition of acids. The salts of this acid saturate 2 atoms of base; on the addition of concentrated sulphuric acid, sulphurous acid is developed.

*Thionuric acid* may be regarded as a combination of 1 atom of alloxan with 1 atom of ammonia and 2 atoms of sulphurous acid, for  $C_8H_4N_2O_{10} + H_3N + S_2O_4 = C_8H_7N_3S_2O_{14}$ .

*Uramile*,  $C_8H_5N_3O_6$ , is produced either by simply exposing thionuric acid to ebullition, or by treating thionurate of ammonia with an excess of hydrochloric acid; it forms minute, silky, glistening needles, and on exposure to the atmosphere and to warmth, assumes a rose-red tint. It is insoluble in cold water and only dissolves slightly in boiling water; the caustic alkalis and concentrated sulphuric acid dissolve, but do not decompose it: by simple ebullition, however, its solutions become decomposed. The alkaline solution of uramile on exposure to the air assumes a purple-red tint, and deposits green crystals with a metallic lustre.

On simply boiling *thionuric acid*, 2 atoms of *sulphuric acid* are given off, and *uramile* is formed; for  $C_8H_7N_3S_2O_{14} - 2SO_3.HO = C_8H_5N_3O_6$ .

*Uramile* may be regarded as uric acid in which the urea is replaced by 1 atom of ammonia and 2 atoms of water; it is, therefore, hypothetically composed of 1 atom of urilic acid, 1 atom of ammonia, and 2 atoms of water, for  $C_8N_2O_4 + H_3N + 2HO = C_8H_5N_3O_6$ .

*Uramilic acid*,  $C_{16}H_{10}N_5O_{15}$ , is formed by boiling uramile either with

a solution of potash or with dilute acids; it crystallizes in colorless, four-sided prisms, or silky, glistening needles, is soluble in water, faintly reddens litmus, dissolves without the development of gas, or the communication of color, in sulphuric acid; is decomposed by nitric acid, and forms soluble salts only with the alkalis.

Acids and alkalis expel 1 atom of ammonia from 2 atoms of uramile, which, in its place, receive 3 atoms of water;  $C_{16}H_{10}N_6O_{12} - H_3N + 3HO = C_{16}H_{10}N_5O_{15}$ .

*Alloxantin*,  $C_8H_5N_2O_{10}$ , is formed by boiling 1 part of uric acid with 32 parts of water, then gradually adding dilute nitric acid, and finally evaporating the fluid to one-third of its volume: after some time crystals of alloxantin separate themselves. It is prepared from alloxan by the action of reducing bodies, as for instance, sulphuretted hydrogen, or hydrochloric acid and zinc. It crystallizes in oblique four-sided prisms, which at first are colorless, but on exposure to the air become yellowish, and if acted on by the vapor of ammonia, become red. It is slightly soluble in cold, but dissolves readily in hot water, it reddens litmus, and is converted by chlorine into alloxan; with baryta-water it gives a violet-colored precipitate.

When very dilute nitric acid acts on uric acid, the urilic acid takes up 1 atom of oxygen from the nitric acid, and 5 atoms of water in order to form alloxantin ( $C_8N_2O_4 + O + 5HO = C_8H_5N_2O_{10}$ ), while the hyponitric acid which is formed, becoming decomposed into nitrous and nitric acids, partly combines with and partly decomposes the urea of the uric acid.

On treating alloxan with sulphuretted hydrogen, the sulphur separates, while the hydrogen unites with the alloxan, and forms alloxantin,  $C_8H_4N_2O_{10} + H = C_8H_5N_2O_{10}$ .

*Murexide*,  $C_{12}H_6N_5O_8$ , *purpurate of ammonia*, may be obtained by several very different methods. The most simple means of preparing it is by boiling equal parts of uramile and red oxide of mercury with 40 parts of water and a very small quantity of ammonia; the purple-red fluid which is thus obtained must be filtered, and after standing some time will deposit crystals of murexide. This body may also be prepared by dissolving uric acid in dilute nitric acid, and evaporating the fluid till it assumes a reddish tint; after it has cooled to  $70^\circ$  it must be saturated with dilute ammonia, diluted with half its volume of water, and allowed to stand.

*Murexide* crystallizes in short four-sided prisms, two of whose surfaces present a cantharides-green, glistening appearance: in refracted light these crystals present a garnet-red tint; when pulverized it is of a brownish-red color, and under the burnishing rod, presents a green, metallic lustre; it is insoluble in alcohol and ether, slightly soluble in cold, but freely in hot water, and it dissolves in a solution of potash, communicating an indigo-blue color to the fluid. It is decomposed by all the mineral acids.

In the preparation of *murexide* from *uramile* and *red oxide of mercury*, 2 atoms of uramile take up 3 atoms of oxygen from the mercury, and form 1 atom of *murexide*, 1 atom of *alloxanic acid* and 3 atoms of *water* ( $C_{16}H_{10}N_6O_{12} + 3O = C_{12}H_6N_5O_8 + C_4HNO_4 + 3HO$ ).

When uric acid is dissolved in dilute nitric acid, the principal product

is alloxantin, which, by the action of the nitric acid during evaporation, is in part converted into alloxan, from which murexide is formed on the addition of ammonia; for 1 atom of *alloxan*, 2 atoms of alloxantin, and 4 atoms of ammonia, yield 2 atoms of *murexide* and 14 atoms of *water* ( $C_8H_4N_2O_{10} + C_{16}H_{10}N_4O_{20} + H_{12}N_4 = C_{24}H_{12}N_{10}O_{16} + H_{14}O_{14}$ ).

*Murexan*,  $C_6H_4N_2O_5$ , *purpuric acid*, is prepared by dissolving murexide in a solution of potash, boiling, and supersaturating with dilute sulphuric acid; it crystallizes in silky, glistening scales, is insoluble in water and in dilute acids, but dissolves unchanged in concentrated sulphuric acid; it likewise dissolves in the alkalies, without, however, neutralizing them.

On treating murexide with alkalies or with acids, 2 atoms of *murexide* take up 11 atoms of *water*, and are converted into 1 atom of *alloxan*, 1 atom of *alloxantin*, 1 atom of *murexan*, 1 atom of *urea*, and 2 atoms of *ammonia* ( $C_{24}H_{12}N_{10}O_{16} + H_{11}O_{11} = C_8H_4N_2O_{10} + C_8H_5N_2O_{10} + C_6H_4N_2O_5 + C_2H_4N_2O_2 + H_6N_2$ ).

*Preparation*—The best method of preparing uric acid is that given by Bensch. The excrements of serpents or birds, or calculi of uric acid, are boiled in a solution of 1 part of hydrate of potash in 20 parts of water till ammoniacal fumes cease to be evolved. A current of carbonic acid is now passed through the solution till the fluid almost ceases to have any alkaline reaction; the precipitated urate of potash is washed with cold water till it begins to dissolve; on now dissolving this potash-salt in a solution of potash, warming it, and pouring it into an excess of warmed hydrochloric acid, we obtain a precipitate of pure uric acid.

*Tests*.—Uric acid possesses such characteristic properties, and differs in so many respects from all other substances occurring in the animal body, that it can hardly be confounded with any other substance, unless possibly with xanthine and guanine (see p. 156 and p. 159); and from these it may be distinguished with extreme readiness and certainty, by the relation of its alkaline salts towards carbonic acid and the alkaline bicarbonates. Uric acid is, however, principally distinguished from all other organic substances (except perhaps from caffeine) by the murexide test, that is to say, by the purplish-red residue which its solution in nitric acid leaves on evaporation; the further addition of caustic potash should, however, never be omitted, by which a yet more distinct reaction—the development of a splendid violet tint—is induced.

All chemical means would, however, frequently fail, and the presence of uric acid would remain undetected, where the quantity of matter to be examined is so small as to afford very slight traces of uric acid, if we were not in possession of the microscope, whose use in physiological chemistry is inestimable. No substance presents such characteristic and so easily determinable crystalline forms under the microscope as uric acid, or crystallizes so readily. Hence it may be detected with ease and certainty by all who are moderately familiar with the use of the microscope, and with the various forms which the crystals of uric acid present. Although, to beginners, the form of the crystals of uric acid appears truly protean, yet with some knowledge of crystallography one form may very readily be deduced from another. We must, however, here

refer to the admirable analysis of the crystallogenes and crystallography of uric acid, as given by Schmidt.<sup>1</sup> For those who are acquainted with crystallography, it is sufficient to give the symbols for the perfect combination of the crystal of uric acid:  $\infty\bar{P}2. \infty P. \infty\bar{P}2. \infty\bar{P} \infty. 0P.$

For the benefit of those who are unlearned in crystallography, we will remark that uric acid when it gradually and spontaneously separates from urine, appears in most cases in the whet-stone form, that is to say, it forms flat tablets, which resemble sections made with the double knife through strongly bi-convex lenses or rhombic tablets, whose obtuse angles have been rounded. As the urinary pigment adheres very tenaciously to the uric acid, it is only rarely that these crystals are devoid of color; and if we see a crystal presenting an extraordinary form and of a yellow color, the probability is that it is a crystal of uric acid. On artificially separating uric acid from its salts it often appears in perfect rhombic tablets, and even oftener in six-sided plates (resembling those of cystine); when uric acid crystallizes very slowly it forms elongated rectangular tablets or parallelopipeds, or rectangular four-sided prisms, with horizontal terminal planes; the latter are often grouped together in clusters; we also have barrel-shaped or cylindrical prisms, which are composed of the more rarely occurring elliptic tablets; and finally saw-like or toothed crystals, and many derivatives of these forms. If we cannot decide with certainty regarding the presence of uric acid from the form of a crystal, we must dissolve it in potash, place it under the microscope, and add a minute drop of acetic acid; we shall then always obtain one of the more common forms.

Fig. 13.



Uric acid.

Fig. 14.



Uric Acid.

A quantitative determination of the uric acid<sup>2</sup> in urine is best made from the residue not taken up by alcohol; by simply treating it with dilute hydrochloric acid, the earths, &c., are got rid of, and nothing but uric acid and mucus remains; their separation may be effected by dissolving them in a dilute solution of potash, from which the uric acid

<sup>1</sup> Entwurf, u. s. w. S. 28-34.

<sup>2</sup> Journ. f. pr. Ch. Bd. 25, S. 17.



may be precipitated by acetic or hydrochloric acid. The pigment adhering to the uric acid exercises no appreciable influence on the quantitative determination of this substance (Heintz).<sup>1</sup>

To institute a quantitative determination of the uric acid in the blood or any other albuminous fluid is a more difficult and far more precarious operation. For this purpose we take the clear serum and evaporate it to dryness, without previously removing the coagulated albumen by filtration; for if we filtered, the whole process would be very prolonged, as the coagulated serum would become little more than a solid mass of moist coagula, whose thorough washing, even by the addition of much water, would be impossible (see the observations in a future page "on the quantitative determination of albumen"). We now extract the solid residue of the serum with alcohol, and afterwards with hot water; as the uric acid in alkaline fluids, and consequently also in the serum, must be combined with an alkali, it is in the aqueous extract that we must always search for it; during the evaporation of the aqueous extract, membranes usually form on the surface of the fluid, which must be removed, but whose removal must slightly affect the accuracy of the analysis; when the aqueous extract has been concentrated to a very small volume, it must be treated with an excess of acetic acid. The uric acid, if its quantity be small, separates very gradually, and unless the acetic acid has been added in great excess, it is usually accompanied with the deposition of a little protein-compound, of whose presence among the crystals of uric acid we can readily convince ourselves by the microscope. It must then be passed through a filter, whose weight has been previously ascertained; and, after careful drying, must be weighed. When the blood is examined qualitatively for uric acid, we must proceed in precisely the same way.

#### *Physiological Relations.*

*Occurrence.*—Uric acid always occurs in the *urine of healthy men*, in the ratio of about one to a thousand parts of urine, as appears from the mean of numerous experiments instituted under different conditions. While living on a mixed diet, the average amount of uric acid which I excreted in 24 hours was 1.183 grammes; according to Becquerel's observations made on 8 different persons, the quantity excreted by healthy men in 24 hours, did not amount to more than from 0.495 to 0.557 of a gramme.

I regret that I must here remark, that the laborious analyses which I made of my own urine cannot altogether serve as standards of comparison for other urines, as when I instituted those observations I was affected with softening of the tissue of the left lung.

Uric acid also occurs in the urine of *carnivorous mammalia*, although generally in far less quantity than in that of man. In the urine of *omnivora*, as, for instance, in that of the pig, neither Boussingault<sup>2</sup> nor Von Bibra<sup>3</sup> succeeded in detecting uric acid. In the urine of *graminivorous mammalia* this acid has never been found, except by Brücke<sup>4</sup> [and by Fownes:<sup>5</sup> G. E. D.], although according to Wöhler it occurs in

<sup>1</sup> Möller's Archiv. 1846, S. 383-389.

<sup>2</sup> Ann. de Chim. et de Phys. 3 Sér. T. 15, pp. 97-114.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 98-112.

<sup>4</sup> Journ. f. pr. Ch. Bd. 25, S. 254.

<sup>5</sup> Phil. Mag. vol. 21, p. 383.

considerable quantity in the urine of calves, while still sucking (compare p. 179). The peculiar urine of birds, both carnivorous and granivorous, and of serpents (which, as is well-known, is generally discharged with the solid excrements, although in snakes it is often unmixed with the latter), consists almost entirely of urates. In the urine of *tortoises* uric acid has been found by Marchand<sup>1</sup> and myself, and Taylor<sup>2</sup> has discovered it in urinary calculi from the *Iguana*. That the red excrement of *butterflies* consists essentially of alkaline urates, and that the excrement of many *beetles* contains the same substances, has been long known; I have, however, not only found uric acid in the excrements of many *larvæ*,<sup>3</sup> but also in large quantities in those vessels of larvæ, to which comparative anatomists have applied the name of biliary vessels.

It is well known that the substance called guano is produced from the excrements of sea-birds; and that it is found not only in the islands of the South Sea (especially in the neighborhood of Chili), but also on the coast of Africa and even in England.

In the urine of the lion, Hieronymi<sup>4</sup> found only 0.022% of uric acid, and Vauquelin could find none whatever.

The *nature of the food* exerts far less influence on the amount of the uric acid which is secreted than on that of the urica. While living on a mixed diet I<sup>5</sup> discharged on an average 1.1 gramme of uric acid in 24 hours, while during a strictly animal and a strictly vegetable diet, the respective amounts were 1.4 and 1.0 grammes.

As the activity of the skin can to a certain degree replace that of the kidneys, it is easy to understand how an increased activity of the skin may cause a diminution of the uric acid in the urine; hence it was that Fourcroy<sup>6</sup> found that the urine of a man contained more uric acid in winter than in summer, and that Marcet<sup>7</sup> was led to the conclusion that the uric acid diminishes in the urine after severe perspiration. Schultens<sup>8</sup> found that in Holland, where, in consequence of the great humidity of the atmosphere, the cutaneous transpiration is diminished, the amount of uric acid varied from 0.21 to 0.67%; for a similar reason, in tropical countries, lithiasis is altogether unknown. These observations, however, merely show it is impossible to lay down numerically any general standard of comparison.

Generally, I have only examined the morning urine, in which I have even found as much as 0.878% of uric acid; investigations regarding the relative qualities of the excreted urinary constituents, can only lead to any useful results when they are instituted on one and the same person, and on the whole urine passed in 24 hours for several days in succession. I have endeavored to arrive at results, in accordance with the above principles, respecting the amount of urine discharged under different conditions, but I have failed in discovering anything further than *that in winter more water is certainly discharged through the urinary blad-*

<sup>1</sup> Journ. f. pr. Ch. Bd. 35, S. 244-247.

<sup>3</sup> Jahresb. d. phys. Ch. 1844, S. 25.

<sup>5</sup> Journ. f. pr. Ch. Bd. 25, S. 254.

<sup>7</sup> An Essay on Calculous Disorders, 1817, p. 176.

<sup>8</sup> N. Gehlen's Journ. Bd. 3, S. 4.

<sup>2</sup> Phil. Mag. vol. 28, pp. 36-46.

<sup>4</sup> Jahrb. de Ch. u. Phys. Bd. 8, S. 822.

<sup>6</sup> Syst. de Connaiss. chim. T. 10, p. 236.

der, but that in summer, during continuous perspirations, the solid constituents, and especially the uric acid, are neither more nor less than in winter. It is unnecessary to give the numerical results from which these conclusions were drawn.

There are however other conditions which give rise both to an absolute and a relative augmentation of the uric acid on the urine, and in the first place amongst them we must notice *disturbed or imperfect digestion*.

Thus, I have observed both in myself and in several other persons, that if indigestible food or spirituous liquors not sufficiently spiced be taken shortly before bed-time, the morning urine always deposited a considerable sediment. While in the normal state the ratio of the uric acid to the uræa = 1 : 28 to 30, I found that in urine passed after indigestion, the ratio = 1 : 23 to 26, and that the ratio of the uric acid to the other solid constituents, which is ordinarily about = 1 : 60 was now = 1 : 41 to 52, so that the amount of uric acid is here not only increased at the expense of the uræa, but also at that of the other solid constituents of the urine. In the most marked case, I found in 100 parts of solid residue 2.4 of uric acid, 35.2 of uræa, and 62.4 of other solid constituents : hence the latter were absolutely increased in this urine.

Consequently it is easy to understand why there is an augmentation of the uric acid in the urine, in many of those cases which the older physicians regarded as *stases* of the portal circulation, hæmorrhoids, and arthritis.

An augmentation in the amount of uric acid in the urine always accompanies the group of symptoms which we are in the habit of designating as fever, the uric acid either separating or remaining dissolved ; for no conclusions can be drawn regarding the quality of uric acid in a specimen of urine, from the formation of a sediment.

I can fully confirm Becquerel's<sup>1</sup> observations on this point by my own experience.

The sediment which is deposited from acid urine in fever, and in almost all diseases accompanied with severe fever, has long been misunderstood in reference to its chemical composition. Originally it was regarded as a precipitate of amorphous uric acid, and subsequently (and almost to the present time) it was regarded as urate of ammonia. It has, however, been fully demonstrated both by myself<sup>2</sup> and Heintz,<sup>3</sup> that this sediment consists of urate of soda mixed with very small quantities of urate of lime and urate of ammonia. It may be very easily and quickly distinguished from any other urinary sediment, both by the microscope and by the application of a gentle warmth : under the microscope it certainly shows little that is characteristic ; it forms fine granules which are sometimes aggregated in irregular heaps, sometimes conglomerated so as to resemble granular cells, and in some instances uniformly distributed over the field of the microscope : as the characteristic forms of uric acid almost immediately appear on the addition of a stronger acid, it is impossible that it can be confounded with any other urinary sediment. An even more simple method of ascertaining that this sedi-

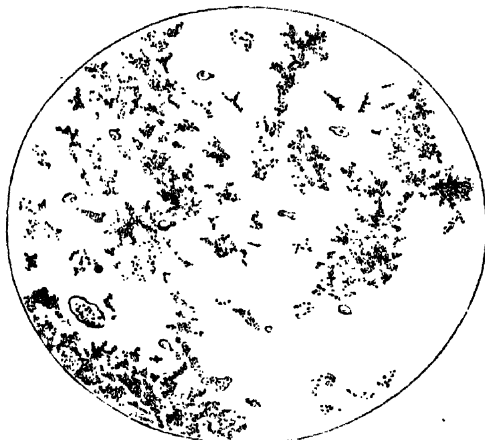
<sup>1</sup> *Séméiotique des Urines*, pp. 51 and 249, or pp. 40-50 and 126-180 of the German Translation.

<sup>2</sup> *Jahresber. d. phys. Ch.* 1844, S. 26.

<sup>3</sup> *Müller's Arch.* 1845, S. 230-261.

ment consists of urate of soda, is afforded by the circumstance that it dissolves at  $50^{\circ}$ , so that urine rendered turbid by it, when raised to that temperature, becomes clear and limpid.

Fig. 15.



Urate of Soda deposited from Urine.

It would be both superfluous and wearisome to recapitulate the arguments adduced by Becquerel, myself, and Heintz, against the opinion of Bird, who maintains that this sediment is always urate of ammonia, as the actual nature of the deposit has been so completely established. I will here only remark that, as I long ago found, and as Liebig has since confirmed, scarcely any ammonia occurs in urine, and that, according to the direct analysis of the sediment made by Heintz, scarcely 1% of ammonia could be found in it.

Much has also been written to prove that uric acid does not exist free in the urine, but in a state of combination with alkalis; but it requires only a moderate knowledge of the properties of uric acid and its salts to perceive that there is nothing wonderful in the presence of an acid urate in an acid fluid, and that the occurrence of acid urate of soda is perfectly natural. Ure<sup>1</sup> and Lipowitz<sup>2</sup> were the first to direct attention to the circumstance which was afterwards very prominently brought forward by Liebig, that phosphate of soda might be one of the solvents of uric acid, and that thus an acid urate of soda and an acid phosphate of soda might be produced. Berzelius,<sup>3</sup> however, has remarked that there are very few solutions of alkaline salts in which uric acid does not dissolve more readily than in water, and that it, for the most part, separates from these solutions as uric acid, and not as an acid alkali-salt. I have, however, especially remarked (*Op. cit.*) that uric acid may extract soda from alkaline lactates, and from compounds of the alkalis with other organic acids, and that the acid salt thus formed communicates an acid reaction to the previously neutral fluid; the urate of soda then separates from a pure mixture in a crystalline form, but from a solution containing extractive matter, as the urine, in an amorphous state, and dissolves again very readily when heated to  $50^{\circ}$ .

<sup>1</sup> Medical Gazette, vol. 35, p. 188.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 38, S. 350.

<sup>3</sup> Jahresber. Bd. 26, S. 873.

The appearance of this sediment of urate of soda (Prout's amorphous and impalpable yellow sediment) is by no means to be regarded as a pathological symptom: it is nothing more than an augmentation of a salt normally existing in the urine, induced by simple physiological relations. Hence we especially observe the formation of such sediments, when, for any reason, the due interchange of the gases in the lungs does not take place, or when, from disturbances of the circulation, the blood does not really permeate the pulmonary vessels. Thus a sediment of this nature may be noticed in men and animals when there is an insufficiency of proper exercise; carnivorous animals, which in their natural state secrete so little uric acid, after long confinement frequently pass a sedimentary urine, especially when they have been reared in cages, and have been attacked by osteomalacia. In fully developed emphysema, or even when only a part of the lung has lost some of its elasticity, a sedimentary urine is one of the most common symptoms. Heart-diseases, enlargements of the liver, &c., are associated with disturbances of the circulation, and hence give rise to a sedimentary urine. It is to such diseases as these that illogical, ontological names—such as hæmorrhoids, gout, &c.—have been applied. Large masses of secreted urate of soda are found in no disease, except in the true granular liver, which obviously can never exist without considerable disturbance of the circulation. In fever also, the due relation between respiration and circulation is no longer maintained, and hence there is an augmentation of the uric acid in the urine; for none but mere chemists could be led to the erroneous idea, that in fever too much oxygen is conveyed to the blood—in short that fever is attended by too rapid a process of oxidation. \* Becquerel's extended observations on urine in diseases, may be profitably compared with the above results of my own experience.

Bird<sup>1</sup> and many others maintain that in gout there is an increased secretion of uric acid: my own experience, however, perfectly confirms that of Garrod,<sup>2</sup> who found that there was a constant and well-marked diminution of the uric acid in the urine before the paroxysm in acute gout, and always in chronic gout (a term which applies only to those cases in which the disease is accompanied by depositions in the joints); while, on the other hand, in rheumatism, especially in acute articular rheumatism, the amount of uric acid in the urine is very much increased—a point in which all observers coincide.

It is extremely seldom that *free uric acid* is found in freshly discharged urine, and its presence there may generally be regarded as a sign of some extremely severe morbid process.

I have *never* been able to find separated crystals of uric acid in urine immediately after its emission, although they may often be found when it has stood for an hour or more. In the great majority of cases the uric acid is formed from the urate of soda after the exposure of the urine to the atmosphere, by the process of acid urinary fermentation which has been so carefully studied by J. Scherer.<sup>3</sup>

Healthy and febrile urine only differ in this point, that the one contains additional elements by which the formation of lactic acid is excited

<sup>1</sup> Urinary Deposits, 3d edit. p. 134.

<sup>2</sup> Medico-Chir. Trans. Vol. 31, p. 86.

<sup>3</sup> Untersuch, S. 1-17.

and promoted. We shall return on a future occasion to this beautiful investigation of Scherer's. I have never seen free uric acid discharged directly from the bladder with the urine except in cases of the calculous diathesis or of pre-existing gravel.

Even in alkaline urine it is very seldom that *urate of ammonia* occurs as a sediment; in these cases it is found in white opaque granules, which as has been already stated, when seen under the microscope, appear as dark globules, studded with a few acicular crystals. It scarcely ever occurs except in urine which, by long exposure to the air, has undergone the alkaline fermentation. Even in the alkaline urine of patients with paralysis of the bladder dependent on spinal disease, it is very rarely that I have found these clusters of urate of ammonia. In the alkaline urine that is sometimes passed in other conditions of the system, it is never found.

Uric acid, like urea, also exists in the *blood*; it has been found there in healthy as well as in diseased states, and especially after extirpation of the kidneys by Strahl and Lieberkühn,<sup>1</sup> as well as recently by Garrod,<sup>2</sup> who observes that in arthritis (but not in acute articular rheumatism), it is invariably, and in Bright's disease it is very often increased in the blood.

My own observations for the most part confirm those of Garrod. I first happened to convince myself of the presence of uric acid in the blood of carnivora in examining the blood of a very large mastiff who died in consequence of an artificial gastric fistula which I had established. The serum was freed from its albumen by boiling and with the aid of acetic acid; the strongly evaporated filtered fluid was extracted with alcohol in order that urea might be sought for; the residue, insoluble in alcohol, exhibited, under the microscope, most unquestionable crystals of uric acid; my attention being thus drawn to the subject, I examined the blood of two other dogs by the same mode of analysis, and convinced myself of the presence of uric acid, not only by the microscope, but also by the murexide test. Garrod asserts that he has often found uric acid in the blood of healthy men, while Strahl and Lieberkühn failed equally in detecting it in the blood of men and of birds; once only they found uric acid in the blood of a dog; they recognized it however with great distinctness, and on many occasions, in the blood of frogs, dogs, and cats, after the extirpation of the kidneys. Garrod found 0.005%, 0.004%, and in one case, even 0.0175% of uric acid in the serum of the blood of gouty patients. In acute articular rheumatism he could only discover traces of uric acid in the blood; in Bright's disease the uric acid of the blood occurred in very variable quantities (from 100 parts of serum he obtained the following quantities, 0.0037, 0.0055, 0.0012, and 0.0027 parts.)

In Germany we have few opportunities of repeating Garrod's experiments regarding the quantity of uric acid in the blood of gouty patients, for in this country we should certainly hesitate before abstracting such masses of blood as he employed in his analyses; he never operated on less than two pounds of blood.

<sup>1</sup> Harnsaure im Blut, u. s. w. Berlin, 1848.

<sup>2</sup> Medico-Chir. Trans. Vol. 31, pp. 87-92.

Scherer<sup>1</sup> has found uric acid in considerable quantity, as a normal constituent, in the juice of the spleen. [Mr. Henry Gray (see his prize Essay "On the Structure and Use of the Spleen," London, 1854, p. 209), being anxious to confirm the observations of Scherer regarding the presence of uric acid and hypoxanthine in the spleen-pulp, worked in one experiment on the spleens of twenty-five oxen, but wholly failed in detecting either of these substances; nor was he more successful with human spleens.—G. E. D.]

Urate of soda is very often found in gouty nodules or concretions, as is shown by the analyses of Wollaston, Laugier, Wurzer, Pauquy, and Bor. My own limited observations entirely accord with the statements of these chemists. The concretions form, for the most part, yellowish white, soft masses, speckled here and there with red spots; on exposure to the atmosphere they harden; examined under the microscope they present the most beautiful tufts of crystals of urate of soda.

Wolf<sup>2</sup> asserts that he discovered uric acid in the sweat of arthritic patients; I have made many attempts to detect it in such cases, but have never yet been successful.

Unfortunately the idea of *gout* in medicine is so vague that it would be well, if, for the present, it were altogether expelled from science. The pathologists are wont to refer to the chemist for the elucidation of this singular disease, but they should rather consider that it is their place to furnish the chemist with more exact ideas regarding this mysterious affection before seeking for an explanation. It must, moreover, be observed that, notwithstanding their assertions to the contrary, pathologists have not yet taught us to distinguish any appreciable difference between gout and rheumatism; while we find from pathological anatomy that the group of symptoms which has generally been regarded as characteristic of the former of these diseases may yield very different results in reference to alterations in the tissues as revealed after death. We most commonly meet with diseases of the osseous system, with osteomalacia in young persons and adults, an affection in which the bones become poorer in earths, and consequently more flexible, than in their natural state, or with osteoporosis or osteospathyrosis, where there is resorption of the cartilage as well as of the earths, as resulting from gout: but the essential principle of the disease cannot lie in this resorption, since often in one and the same bone we find sclerosis and porosis; the change which the bone undergoes is solely dependent on the nature of the exudation which is thrown out; if the latter be very consistent (fibrinous?) it puts on an appearance of callus, deposits an excess of bone-earth, and the affected part becomes sclerotic; if, on the other hand, it be fluid, resorption takes place, and the result is osteoporosis; if it exhibit a tendency to decomposition and become ichorous, caries as well as pyæmia may ensue. Unfortunately, however, these alterations in the osseous system are not peculiar to gout, but occur both from purely local causes, and from other general diseases, especially from syphilis. The diseased condition of the osseous system, however constantly it may be observed in gout, when we adhere to the strictest definition of the term,

<sup>1</sup> Verhandl. d. phys.-med. Ges. zu Würzburg. Bd. 2, S. 299.

<sup>2</sup> Diss. sist. casum Calculositatis. Tub. 1817.

affords us no firm starting-point ; we must, consequently, have recourse to the nodules and concretions, but these earthy deposits may exist independently of gout, and there remains no characteristic of the nature of gout excepting the concretions of urate of soda ; yet how seldom do even these occur ; and how far advanced must be the malady before we can base our diagnosis on their presence ! The accumulation of great quantities of uric acid in the blood, independently of other symptoms, is also devoid of importance, since, according to Garrod, this may likewise occur in Bright's disease. In a word, we know not the nature of arthritis ; and if this ever be elucidated by physiologico-chemical investigations, I believe that the sole method which will conduce to this end will be that of ascertaining the relation in which the chemical constitution of the blood and urine stands to the above-named diseases of the osseous system, and to osteomalacia in particular.

It seems to us still more inappropriate and still less in accordance with a rational natural inquiry, if, basing our views on a preconceived and misunderstood proposition, we philosophize on the analogy of "gout, gravel, and stone ;" *a priori* explanations of morbid processes such as have been attempted in the organico-chemical department of medicine, have usually failed in yielding any results, from the misconception that, without physiology and pathological anatomy, medicine might be established in accordance with subjective chemical views. The pretended oxidation of the constituents of the blood, which was supposed to explain phthisis as well as gout and stone, is not the simple method by which alone specific diseases or individual well-characterized processes can be explained with scientific accuracy. For *there are no acute and but few chronic diseases in which the oxidation of the constituents of the blood is not diminished or impeded.* The proof of the assertion will, in a future part of this work, be made as evident as the fact *that there is no disease characterized by a too sudden or rapid oxidation of the blood.*

*Origin.*—Since we have already (see p. 156) mentioned that urea is in part derived from uric acid, there can be no doubt that the latter, like the former, must rank amongst the excrementitious matters. Although we have no numerical proof that in human urine the urea stands in an inverse ratio to the uric acid, that is to say, that with an augmentation of the uric acid there is a corresponding diminution of the urea, yet the numerical results of Becquerel and others show that there is at least such an approximate ratio. The recent experiments of Wöhler and Frerichs,<sup>1</sup> in which the introduction of uric acid into the organism by the *primæ viæ* or by the veins, was followed by an augmentation of the urea and oxalate of lime in the urine, afford tolerably strong evidence that the uric acid in the animal organism undergoes a decomposition into urea and oxalic acid precisely similar to that which can be artificially induced by peroxide of lead. Now, if the urea be produced from the uric acid by the partial oxidation of the latter, anything impeding this process must cause less urea and more uric acid to be separated by the kidneys, and hence we see why the amount of uric acid in the urine must be increased in fevers and other disturbances in the circulation and respiration ; we have seen that in like states oxalate

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 338–342.



of lime and lactic acid increase for a precisely similar reason, and without wishing to introduce rude chemical views into the science of general life, nothing seems more simple, and in accordance with nature, than this explanation of the origin and augmentation of uric acid. We regard uric acid as a substance which stands one degree higher in the scale of the descending metamorphosis of matter than urea. The present condition of science does not admit of our specially indicating the substances from which it is first produced, or the locality in which it is formed.

Sediments of urate of soda are commonly ranked amongst the critical discharges. A rational system of medicine can no longer, in accordance with the doctrines of Hippocrates, regard these excretions as true crises of diseases, but must rather consider them only as incidental symptoms, or as necessary consequences of certain processes. In the present day we regard the crises merely as very abundant eliminations of excrementitious matters which must occur when the substances rendered effete during the fever, and which have accumulated in the blood while the functions of the excreting organs were more or less impeded, are fit for simultaneous secretion, and are thus given off to the outer world by their ordinary channels.

### INOSIC ACID.— $C_{10}H_6N_2O_{10} \cdot HO$ .

#### *Chemical Relations.*

*Properties.*—This acid is not crystallizable; it forms a syrupy fluid, which is converted by alcohol into a solid, hard mass; it dissolves readily in water, but is insoluble in alcohol and ether; it reddens litmus strongly, possesses an agreeable taste of the juice of meat, is decomposed by heating, and in part, if its solution be boiled.

*Composition.*—According to the above formula, which Liebig,<sup>1</sup> the discoverer of this acid, deduced from his analysis of the baryta-salt, this acid consists of:

Carbon, . . . . .	10 atoms,	. . . . .	32.787
Hydrogen, . . . . .	6 "	. . . . .	3.279
Nitrogen, . . . . .	2 "	. . . . .	15.800
Oxygen, . . . . .	10 "	. . . . .	48.716
Water, . . . . .	1 "	. . . . .	4.918
			100.000

The atomic weight of the hypothetical anhydrous acid = 2175.0, and its saturating capacity = 4.597. This acid is unquestionably no simple oxide of a ternary radical, but contains certain proximate constituents; its products of metamorphosis have, however, as yet been so little studied that we cannot even form any conjecture regarding the adjunct or the peculiar acid contained in it. Liebig remarks that it may be regarded as composed of 1 equivalent of acetic acid, 2 equivalents of oxalic acid, and 1 equivalent of urea.

*Combinations.*—The alkaline inosates are soluble in water, are crys-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 325-335.

tallizable, and, when heated on a platinum spatula, diffuse a powerful and agreeable odor of roasted meat.

*Inosate of potash*,  $\text{KO} \cdot \text{C}_{10}\text{H}_6\text{N}_2\text{O}_{10} + 7\text{HO}$ , occurs in long, delicate, four-sided prisms; on the addition of alcohol to a concentrated aqueous solution, this salt separates in fine nacreous scales.

*Inosate of baryta*,  $\text{BaO} \cdot \text{C}_{10}\text{H}_6\text{N}_2\text{O}_{10} + 7\text{HO}$ , crystallizes in long four-sided scales of a nacreous lustre, which, when dry, have the aspect of polished silver; it effloresces readily, dissolves freely in hot, very slightly in cold water, and not at all in alcohol. If a solution, saturated at  $70^\circ$ , be heated to boiling, a part of the salt is deposited in the form of a resinous mass.

*Inosate of copper* forms a light blue, amorphous powder, insoluble even in acetic acid.

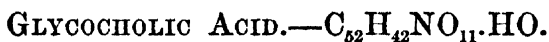
*Inosate of silver* is amorphous, white, and slightly soluble in pure water.

*Preparation*.—If the mother-liquid of the juice of flesh, after the creatine has crystallized and been removed (see p. 128,) be gradually treated with alcohol till the whole become milky, it deposits, in the course of a few days, yellow or white granular, foliated, or acicular crystals of the inosates of potash and baryta, mixed with creatine. Chloride of barium must be added to the hot aqueous solution of these crystals; on cooling there is a deposition of crystals of inosate of baryta, which, by recrystallization, are rendered perfectly pure. By decomposing this salt with sulphuric acid, or the copper-salt with sulphuretted hydrogen, the acid is obtained in a state of purity.

*Tests*.—So little is yet known regarding the properties of this acid, that the only test we can rely upon is the ultimate analysis.

#### *Physiological Relations.*

Liebig has hitherto only found this acid in the fluid of flesh. The few facts which we at present possess regarding this acid throw no light on its mode of formation. From the great quantity of oxygen which it contains, it must be regarded as a product of the decomposition of effete tissues.



#### *Chemical Relations.*

*Properties*.—This acid, which has been named, *par excellence*, *bilic* or *cholic acid*, forms extremely delicate needles, which remain unchanged at  $136^\circ$ ; it has a bitterish-sweet taste, dissolves in 120.5 parts of hot, and 303 parts of cold water; is readily soluble in spirit, but only slightly in ether; it does not crystallize on evaporating the alcoholic solution, but separates as a resinous mass; but it crystallizes from the spirituous solution, mixed with water and exposed in the air to gradual evaporation. The aqueous solution of this acid reddens litmus strongly. It dissolves without change in concentrated acetic acid, cold sulphuric acid, and hydrochloric acid.

The aqueous solution of this acid is not precipitated by acids, neutral acetate of lead, corrosive sublimate, or nitrate of silver; in alkalies it dissolves freely, being precipitated from them by acids, in a resinous form; on standing, especially after the addition of a little ether, the resinous precipitate becomes crystalline. A solution of the acid in combination with an alkali yields no precipitate with chloride of barium; but there are precipitates on the addition of the salts of the oxides of lead and copper and peroxide of iron; nitrate of silver, when added to very dilute solutions, yields a gelatinous precipitate, which, on warming, again dissolves, and on cooling gradually assumes a crystalline form. By prolonged boiling with a solution of potash, or still better, with baryta-water, this acid becomes resolved into the non-nitrogenous cholic acid and glycine (see p. 142). When boiled with concentrated sulphuric or hydrochloric acid, it is resolved into choloidic acid and glycine (Strecker).<sup>1</sup>

With sulphuric acid, and either sugar or acetic acid, glycocholic acid yields the same reaction as cholic acid (see p. 117).

If glycocholic acid be submitted to prolonged ebullition in water, it becomes perfectly insoluble, and breaks up into fragments of six-sided tablets. To this modification the name of *paracholic acid* has been applied by Strecker.

*Composition.*—From numerous analyses of glycocholic acid and its salts, Strecker<sup>2</sup> has deduced for it the above formula, according to which it consists of:

Carbon, . . . . .	52 atoms,	. . . . .	67.097
Hydrogen, . . . . .	42 “ . . . . .	. . . . .	9.082
Nitrogen, . . . . .	1 “ . . . . .	. . . . .	3.011
Oxygen, . . . . .	11 “ . . . . .	. . . . .	18.925
Water, . . . . .	1 “ . . . . .	. . . . .	1.985
<hr/>			
			100.000

The atomic weight of the hypothetical anhydrous acid=5700; and its saturating capacity=1.754.

Hardly a doubt can remain that this is a conjugated acid, when we consider, on the one hand, that we are acquainted with another acid (hippuric acid) from which the same nitrogenous body, glycine, may be separated by acids, and that, on the other hand, there is another acid from which the same non-nitrogenous acid, cholic acid, is liberated by acids, another body, taurine, being simultaneously produced (this taurine in the taurocholic acid taking the place of the glycine in the glycocholic acid). In glycocholic acid we cannot, however, consider glycine, as we know it in its isolated state, to be the adjunct of cholic acid, but must rather assume that the true adjunct of cholic acid, as in the case of hippuric acid, undergoes a change during its separation, by which it forms the body known to us as glycine. If, as in hippuric acid, we regard this adjunct as a group of atoms isomeric with fumaramide, the rational formula of glycocholic acid will be  $\text{=C}_4\text{H}_3\text{NO}_2\cdot\text{C}_{48}\text{H}_{39}\text{O}_9\cdot\text{HO}$ .

*Combinations.*—With alkalies and alkaline earths, glycocholic acid forms very soluble salts; its compounds with the oxides of the heavy

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 66, S. 1-43.

<sup>2</sup> Ibid. Bd. 65, S. 1-87.

metals are, however, insoluble; the glycocholate of silver alone being soluble in boiling water.

*Glycocholate of soda*,  $\text{NaO.C}_{52}\text{H}_{42}\text{NO}_{11}$ , separates from its alcoholic solution, on the addition of ether, in large, glistening, white clusters of radiating needles, resembling wavellite; it is not crystallizable from its watery or spirituous solutions; it dissolves very readily both in water and in spirit (1 part dissolving in 2.56 of spirit at  $15^{\circ}$ ); when heated it melts, burns with a smoky flame, and leaves an ash containing cyanides. *Glycocholate of potash* behaves in a similar manner.

*Glycocholate of ammonia*,  $\text{H}_4\text{NO.C}_{52}\text{H}_{42}\text{NO}_{11}$ , occurs in crystals precisely similar to those of the soda-salt, when it is gradually separated from an alcoholic solution by ether; it dissolves readily in water, yields ammonia on boiling, and then has a faintly acid reaction.

*Glycocholate of baryta*,  $\text{BaO.C}_{52}\text{H}_{42}\text{NO}_{11}$ , is amorphous, has a strongly sweet and slightly bitter taste, is soluble in water and in alcohol, and is not decomposed by carbonic acid.

*Preparation.*—This acid occurs in the bile of most animals, but it is best prepared from the bile, of the ox by one of the two following methods. The bile, first carefully dried in the water-bath, and subsequently *in vacuo*, must be extracted with cold absolute alcohol, and ether must be gradually added to the filtered solution, which is thus rendered turbid, and soon deposits a brownish, tough, resinous mass. If the fluid be now only slightly colored, we must decant it from the semi-fluid precipitate into another vessel, and again gradually add ether; the fluid again becomes milky, and deposits more resinous matter; after a time however, glistening star-like tufts of crystals are deposited, which must be washed with alcohol to which a tenth part of ether has been added, and then rapidly placed *in vacuo*, because the crystals, when moist with ether, rapidly deliquesce into a varnish-like mass; after drying they cease to be acted on by the atmosphere. These crystals are a mixture of the glycocholates of potash and soda. On precipitating the aqueous solution of these crystals with acetate of lead, decomposing the precipitate with carbonate of soda, evaporating the solution of glycocholate of soda, re-dissolving it in alcohol, and again (in the same manner as before) crystallizing by means of ether, we obtain a tolerably pure glycocholate of soda, which, when dissolved in water and treated with dilute sulphuric acid, after a time deposits crystals mingled with oily globules. The latter may be removed by washing with water, leaving the glycocholic acid in a state of purity.

The following is a shorter method of obtaining this acid. The yellowish precipitate thrown down by sugar of lead from fresh bile must be extracted with boiling spirit of 85% and sulphuretted hydrogen passed through the solution. If water be added to the filtered fluid and the mixture be allowed to stand for a considerable time, the acid will separate in a crystalline form; in this case, however, it is better to decompose the lead-salt by carbonate of soda, and then to proceed in accordance with the former method.

Crystallized bile, which is a mixture of the glycocholates of potash and soda, was first prepared by Platner.<sup>1</sup>

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 51, S. 105; Journ. f. pr. Ch. Bd. 40, S. 129–133.

*Tests.*—In attempting to determine the amount of bile in an animal fluid, it is always necessary that the albuminous matters, the substances soluble in water only, and the fats, should be as completely as possible separated. We consequently, in the first place, obtain an alcoholic extract of the substance to be investigated, and ascertain by Pettenkofer's test whether any derivative of the bile be present in it. This point being decided, we can only determine whether one of the acids contained in fresh bile—glycocholic or taurocholic acid, or one of their derivatives, cholic or choloidic acid—be present, when we have a considerable amount of matter to work upon. To pursue this inquiry, we must gradually add from 8 to 12 times its volume of ether to the extract obtained by strong alcohol, and allow the mixture to stand for from 24 to 48 hours; by this time the turbidity of the fluid will have disappeared, and a sediment will have formed, which is either flocculent and viscid, so as to adhere to the walls of the vessel (in which case it consists for the most part of albuminous or extractive matter), or is a resinous, semi-fluid, tough mass (alkaline taurocholates or choloidates), or consists of tufts of well-formed crystals of various sizes, visible to the naked eye, and composed either of cholate or glycocholate of soda. It is worthy of remark that even the smallest quantities of the alkaline glycocholates crystallize from their solution in this way. (From a solution of about 0.07 of a gramme of glycocholate of soda in 150 parts of alcohol, I obtained most beautiful crystals of the salt on the addition of 560 grammes of ether.) These crystals must, however, always be examined microscopically, or at all events with a lens, as many other salts (acetate of soda for instance) separate in a crystalline form under this mode of treatment: they form six-sided prisms with a single very oblique plane of truncation, and as their aqueous solution reacts with Pettenkofer's bile-test, no doubt can remain regarding the presence of glycocholic acid. If the crystals be obtained either in a state of purity or surrounded by syrupy matter, we must separate the acid from the alkali by a little sulphuric acid, and extract with ether, in which the conjugated cholic acids as well as choloidic acid are almost insoluble; if the crystallizable cholic or glycocholic acid be thus isolated, we can determine regarding the presence or absence of one or other of them by boiling with a solution of potash, when, if glycocholic acid be present, ammonia is developed; moreover, the cholate of baryta is a crystallizable salt, while the glycocholate of baryta is amorphous. Glycocholic acid resembles choloidic acid in being only slightly soluble in ether; they may, however, generally be distinguished by the crystallizability of the former acid and of its salts from ethereal-alcoholic solutions; the glycocholate of baryta, indeed, resembles the choloidate in being uncrystallizable, but it differs from the latter in being soluble in water. We shall point out the means of distinguishing between glycocholic and taurocholic acids in our observations on the latter acid.

#### *Physiological Relations.*

*Occurrence.*—As far as our investigations have hitherto extended, this acid has been found in the *bile* of all animals, with the exception of the pig. In reference to its occurrence in other parts and fluids of the

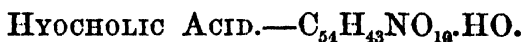
animal body, we have only to repeat what has already been said in pp. 119-20 regarding cholic acid. We meet with such minute quantities of biliary matter in the intestinal canal, in the blood, and in exudations, that until recently they have been, for the most part, entirely overlooked, and it is only by means of Pettenkofer's admirable test that we can now detect them. Important as it would be in a physiological point of view to ascertain whether cholic acid or the conjugated biliary acids occur in the blood, and whether these or choloidic acid occur in the intestine, we must for the present leave these questions altogether undecided.

Kunde, one of my pupils, has very distinctly recognized the presence of biliary matters by means of Pettenkofer's test in the fluid from the hydrocele of an otherwise healthy man. By the same test he was able to demonstrate the presence of biliary matters in the blood of frogs, whose livers he had extirpated. (Of six frogs on which he operated, only two survived.)

*Origin.*—We have already (see p. 120) attempted to show the probability that cholic acid obtains its essential elements from the fats, and that, in short, it is oleic acid conjugated with a non-nitrogenous body. But in glycocholic acid we again meet with the same nitrogenous adjunct which we have already encountered in hippuric acid, and which, consequently, seems to be an ordinary product of decomposition of nitrogenous bodies. We have already remarked (see p. 181) that we are not in a condition to name the proximate source of this adjunct, which is, however, isomeric with fumaramide.

This is not the most appropriate place for entering into the physiological reasons for showing the part which the fat takes in the formation of the principal constituents of the bile, or for balancing the reasons for or against the formation of bile within the hepatic cells. These are subjects pertaining to the second department of our work, in which we shall consider the bile in general as an animal secretion. We may, however, be permitted to remark that the possibility of the primary formation of this acid in the blood is indicated partly by the above-mentioned experiments of Kunde, and partly by the not unfrequent occurrence of icterus independently of any hepatic affection (Virchow), that is to say, without infiltration of the parenchyma of the liver and of the hepatic cells with bile-pigment.

*Uses.*—As we are not at present accurately acquainted with the changes which glycocholic acid undergoes in the intestinal canal, we are unable to state whether this acid exerts any special action in the process of digestion.



#### *Chemical Relations.*

*Properties.*—This acid, discovered and accurately examined by Gundelach and Strecker,<sup>1</sup> forms a white resinous mass, which melts in water at 100° and, like choloidic acid, may be drawn out in long threads; when

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 205-282.

perfectly dry it does not melt at a temperature under  $120^{\circ}$ ; it is only slightly soluble in water, dissolves readily in alcohol, and not at all in ether; it reddens litmus. It dissolves unchanged in cold concentrated nitric and sulphuric acids; but when boiled for some time in either of those acids it yields, like glycocholic acid, glycine and a resinous acid similar to choloidic acid; with concentrated sulphuric acid and either sugar or acetic acid, it yields, like the other biliary acids, a purplish-violet solution; it is only decomposed by a solution of caustic potash, when the mixture is so concentrated as to solidify on cooling. It is unchanged by digestion in moderately concentrated sulphuric acid and peroxide of lead; putrefaction of the bile seems to exert no influence on it; when treated with fuming nitric acid, or decomposed by chromic acid, it yields the same products as choloidic acid, namely *cholesteric acid*, butyric acid, caproic acid, &c.

*Composition.*—According to Gundelach and Strecker, this acid may be obtained in an anhydrous state, so as in its combination with bases to lose no water. From their analyses of the free acid, as well as of its salts, these chemists have deduced the above formula, in accordance with which the free anhydrous acid consists of:

Carbon,	.	.	.	.	.	54 atoms,	.	.	.	.	70.28
Hydrogen,	.	.	.	.	.	43 "	.	.	.	.	9.33
Nitrogen,	.	.	.	.	.	1 "	.	.	.	.	3.04
Oxygen,	.	.	.	.	.	10 "	.	.	.	.	17.35
											<hr/> 100.00

The atomic weight = 5762.5, and its saturating capacity = 1.735.

This acid contains 2 atoms of carbon and 1 atom of hydrogen more, but 1 atom of oxygen less, than glycocholic acid; the fact that, when treated with concentrated mineral acids, it likewise yields glycine, tends to confirm the hypothesis, that hyocholic acid also contains the glycine-yielding adjunct isomeric with fumaramide, and that so much plus of carbon and hydrogen, and minus of oxygen, are respectively added to, and deducted from the non-nitrogenous acid, that the rational formula for this acid would be =  $C_4H_3NO_2 \cdot C_{50}H_{40}O_8$ . But as hyocholic acid when decomposed with nitric acid yields the same volatile fatty acids and cholesteric acid, the non-nitrogenous acid, contained in hyocholic acid, may be presumed to have a constitution analogous to cholic acid (see p. 120), and besides the group of atoms  $C_{12}H_6O_6$  which yields the cholesteric acid ( $C_8H_4O_4$ ) to contain another fluid fatty acid of the formula  $C_nH_{n-3}O_3$  in place of the oleic acid in the cholic acid; and this in point of fact admits of being calculated by subtracting the group of atoms  $C_{12}H_8O_8$  from the hydrate of the non-nitrogenous hyocholoidic acid;  $C_{50}H_{41}O_9 - C_{12}H_8O_6 = C_{38}H_{33}O_3$ , which is exactly the formula of doeglic acid (see p. 111).

That this calculation is a mere fiction is sufficiently obvious, but we believe that such fictions should not be altogether unnoticed, since they stimulate us to further inquiry, even if it were only to determine whether an acid isomeric or identical with doeglic acid existed in the fat of the pig.

*Combinations.*—The *alkaline hyocholates* are not crystallizable; they are soluble in water and alcohol, but not in ether, which completely pre-

precipitates them from their alcoholic solutions. Their taste is bitter without any sweet after-taste, and they redden litmus; like soaps, they are precipitated from their aqueous solutions by alkaline salts, the precipitate containing the base of the salting added in excess; they melt and are inflammable when heated; with the *salts of baryta, lime, and magnesia*, they yield white precipitates soluble when the mixture is raised to the boiling temperature. Their aqueous solutions are precipitated by most of the metallic salts, but their alcoholic solutions are not affected by these reagents. On the addition of an acid to the aqueous solution, the hyocholic acid is entirely precipitated. Neutral acetate of lead yields a white precipitate which does not cake on boiling.

*Hyocholate of potash*,  $\text{KO.C}_{54}\text{H}_{43}\text{NO}_{10}$ , is in its moist state a white amorphous mass which melts in the water-bath, and dissolves as long as it contains either water or spirit. It does not dry at a temperature under  $120^{\circ}$ .

*Hyocholate of Soda*,  $\text{NaO.C}_{54}\text{H}_{43}\text{NO}_{10}$ , forms when dry a brownish mass, which when finely triturated, becomes of a snow-white color; it has a persistent bitter taste without any sweet after-taste. Its solutions are neutral, and are not rendered turbid by carbonic acid. It is precipitated from its alcoholic solution by ether, and from its aqueous solution by soda-salts; it melts when heated, dissolves, and burns with a bright but smoky flame.

*Hyocholate of ammonia*,  $\text{H}_4\text{NO.C}_{54}\text{H}_{43}\text{NO}_{10}$ , is a white crystalline powder. Its solutions become turbid on boiling, and assume an acid reaction. It may be dried over sulphuric acid without loss of ammonia.

*Hyocholate of baryta*,  $\text{BaO.C}_{54}\text{H}_{43}\text{NO}_{10}$ , is a gelatinous substance, freely soluble in spirit, moderately soluble in hot water, and slightly so in cold water.

*Hyocholate of lime*,  $\text{CaO.C}_{54}\text{H}_{43}\text{NO}_{10}$ , is white, amorphous, and rather more soluble in water than the baryta-salt; it is precipitated from its spirituous solution by water and by carbonic acid.

*Hyocholate of lead* is a white powder, which neither cakes when boiled with water nor when dried; it is slightly soluble in water, but freely in spirit, from which it (like all the other salts of this acid) is precipitated by ether. Red litmus is turned blue by the alcoholic solution.

*Hyocholate of silver*,  $\text{AgO.C}_{54}\text{H}_{43}\text{NO}_{10}$ , occurs as a gelatinous precipitate, which, on boiling, becomes flocculent; it dissolves freely in spirit, slightly in cold, but somewhat more easily in hot water.

*Preparation.*—The precipitate caused by the addition of a solution of sulphate of soda to fresh swine's bile is dissolved in absolute alcohol, decolorized by a little animal charcoal, and the soda-salt of the acid precipitated by ether from the alcoholic solution; this is decomposed by dilute sulphuric acid, and the precipitate is dissolved in alcohol, from which the hyocholic acid is thrown down by the addition of water.

*Tests.*—It is only with glycocholic and choloidic acids that this acid can possibly be confounded. From the former it may easily be distinguished by the circumstance that neither it nor its salts can be obtained in a crystalline state by the addition of ether to alcoholic solutions. It is, however, not so readily distinguishable from the latter, because without an elementary analysis, it is impossible to determine its nitrogen; and be



cause, further, when treated with concentrated hydrochloric acid it yields too little glycine to be recognized with certainty, unless, indeed, we have a very large supply of the material to be investigated. The fact that hyocholate of lead neither cakes when dried nor when boiled with water, while the opposite is singularly the case with the glycocholate, affords a tolerably characteristic test. Other differences are for the most part only gradual, and are inapplicable as tests to enable us to distinguish between small quantities of these acids.

### *Physiological Relations.*

This acid has hitherto only been found in the *bile of the pig*, where it exists in combination with soda, potash, and a little ammonia. Our remarks on the origin and uses of glycocholic acid are equally applicable to hyocholic acid.

### TAUROCHOLIC ACID.

#### *Chemical Relations.*

*Properties.*—This acid, which has also been named *choleic acid*, and was formerly known as *bilin*, has not yet been obtained in a state of perfect purity, that is to say, free from glycocholic acid; it cannot be obtained in a crystalline state, and it is more soluble in water than glycocholic acid, while its acid properties are far weaker. It dissolves fats, fatty acids, and cholesterin in large quantities, and is thus the cause why glycocholic acid is not precipitated from fresh ox-bile by acetic or the mineral acids. On exposure to the air, as well as on evaporating a solution of the free acid, decomposition ensues. When boiled with mineral acids it becomes resolved into taurine and choloidic acid; when boiled with alkalis, into taurine, and cholic acid; and when treated with sulphuric acid and sugar, it gives the same reaction as the other essential acids of the bile. The characters of its salts are, however, very distinct from those of the other biliary acids.

*Composition.*—As this acid, like glycocholic acid, becomes resolved, when acted on by mineral acids and by alkalis, into choloidic or cholic acid, while in place of glycine it yields taurine, Strecker,<sup>1</sup> to whom we are especially indebted for our knowledge of this acid and of its properties, correctly argues that its composition is perfectly analogous with that of glycocholic acid, the only difference being that the adjunct in this case is taurine. Abstracting from the formula for taurine 1 atom of water, he assumes that the empirical formula of this acid =  $C_{52}H_{45}NS_2O_{14}$ , and the rational formula =  $C_4H_6NS_2O_5 \cdot C_{48}H_{39}O_9$ . We must therefore regard taurocholic acid as containing an adjunct rich in sulphur, which, on its separation from the cholic acid, becomes converted into taurine, whose properties we have already described at p. 166. By elementary analyses of a mixture of pure alkaline glycocholates and taurocholates, obtained directly from fresh bile, Strecker has further confirmed his view regarding the composition of this acid. Pure taurocholic acid must, there-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 66, S. 43-61.

fore, contain 6.213% of sulphur, while its atomic weight must = 6437.5 and its saturating capacity be 1.553.

*Combinations.*—The *alkaline taurocholates* dissolve readily in water and in alcohol, but are perfectly insoluble in ether; they have no reaction on vegetable colors, and attract water from the atmosphere, but do not deliquesce; when kept for a long time in contact with ether they crystallize; their aqueous solutions have a sweet taste with a bitter after-taste, and do not decompose when evaporated, or when exposed to the air, provided they be pure. These salts when heated melt and burn with a bright smoky flame. Carbonic acid does not decompose their alcoholic solution; their aqueous solution is not precipitated by acids, nor by the alkaline sulphates or chlorides (like the alkaline hyocholates), but by concentrated alkaline solutions; it is not precipitated by the salts of baryta, lime, or magnesia, even on the addition of ammonia, or by neutral acetate of lead; but on the addition of basic acetate of lead, there is a plastery precipitate which dissolves in boiling water, and even more freely in boiling alcohol, and is also soluble in an excess of acetate of lead. Nitrate of silver, even after the addition of ammonia, does not precipitate the taurocholates, neither does corrosive sublimate, but precipitates are induced by nitrate of suboxide of silver, and protochloride of tin. Nitrogenous substances, mucus for instance, set up a process of decomposition in solutions of the alkaline taurocholates, which may be readily ascertained by the circumstance that the solutions then become precipitable by dilute acids. The products which are formed are taurine, alkaline cholates or choloidates, and probably certain combinations of these substances with taurocholic acid that has escaped decomposition. In aqueous solutions of pure alkaline taurocholates, these decompositions are not observed to occur.

*Preparation.*—We have already remarked, that this acid has never yet been prepared in a state of complete purity. In order to separate it as thoroughly as possible from the glycocholic acid which always accompanies it, we in the first place remove from the purified ox-bile the greater part of the glycocholic acid and of the fatty acids by means of neutral acetate of lead, and then precipitate by basic acetate of lead, to which we may add a little ammonia. This precipitate must be decomposed with carbonate of soda, and we must extract the solid residue of the filtered fluid with alcohol. On the addition of ether to the alcoholic solution, a tolerably pure taurocholate of soda is immediately precipitated in the form of a resinous, semifluid, yellow mass. If this be dissolved in a small quantity of water, and all that is precipitable by acetate of silver be thrown down, and if the fluid after filtration be precipitated with basic acetate of lead, and the precipitate, after being thoroughly diffused in a little water, be treated with sulphuretted hydrogen, we obtain tolerably pure taurocholic acid after evaporating *in vacuo*.

*Tests.*—No great weight can be attached to any of the differences in the reaction of the salts of glycocholic and taurocholic acids, when the quantity of the substance presented to us for examination is very small. If, however, we have sufficient material, we must obtain the acids from the alcoholic extract with precisely the same precautions as we have in-

licated in the preceding pages in reference to each of these acids; from the ratio of the precipitate caused by the sugar of lead to that caused by the acetate of lead, we must draw our conclusions regarding the relative quantities of the two acids, and then, by treating the alcoholic solution of the soda-salt with ether, we can determine this point with certainty; indeed, we shall always be most decisively convinced of the presence of taurocholic acid by the exhibition of the taurine, which, even if obtained in only very small quantities, may be recognized with certainty by crystallometric examination under the microscope. Unfortunately, however, the quantities of taurine are so minute, unless when we are acting directly on bile, that it cannot be distinguished and recognized with certainty either by the above means or by its relation towards nitrate of silver and other metallic salts. Nothing further remains for us but to determine the presence of sulphur; having ascertained by Pettenkofer's test that biliary matter is present in the substance under examination, we must extract the spirituous extract with cold absolute alcohol, concentrate this solution, and treat it with ether. A precipitate then falls, which cannot contain any other known sulphurous substance, and which we must fuse and deflagrate with nitrate of potash and caustic potash free from sulphuric acid; if sulphuric acid be found in the residue, we may regard the presence of taurocholic acid as almost certain.

Unfortunately, substances in which it is of interest to detect small quantities of taurocholic acid, are seldom obtained in a state of perfect freshness, and the little taurocholic acid that was originally present is decomposed before we commence our investigations. When we suspect that this acid is present, and have detected biliary matter by Pettenkofer's test in the alcoholic extract, we may hope to find taurine in the aqueous extract, which, however, contains it in such small quantity, and often so intermingled with other substances, that its recognition, even under the microscope, is extremely difficult. We must not attempt to determine the presence of sulphur as a test for taurocholic acid or taurine in the aqueous extract, for this contains both sulphates and other sulphurous organic bodies.

### *Physiological Relations.*

*Occurrence.*—From the determinations of the amount of sulphur, instituted by Bensch<sup>1</sup> and others, we may conclude that taurocholic acid exists not only in the bile of the ox, but in that of the fox, bear, sheep, dog, wolf, goat, and certain birds and fresh-water fish; it has been found in the bile of the frog by Kunde and myself; and that it exists in human bile can hardly be doubted, since, as Gorup-Besanez was the first to prove, taurine may be exhibited from it. It might almost be inferred, from the numerical results obtained by Schlieper<sup>2</sup> in his analysis of the purified bile of a *Boa Anaconda*, that the liver of this serpent secretes taurocholic alone, and none of the other known biliary acids. That this acid is almost entirely absent in the bile of the pig, as shown by the investigations of Strecker, has been already mentioned.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 194–203.

<sup>2</sup> Ibid. Bd. 60, S. 109–112.

Unchanged taurocholic acid has not yet been found in any other animal fluid; but from the experiments of Kunde to which I have already referred (p. 205), it is not improbable that it also occurs in the blood.

*Origin.*—We have very little to say in the present place regarding the production of taurocholic acid: what has been already stated respecting the formation of cholic acid (p. 120), of taurine (p. 168), and of glycocholic acid (p. 205), is equally applicable to the acid under consideration. As it has not yet been found in the blood, it is impossible to decide chemically whether it be primarily formed in the liver from its proximate constituents, or whether it proceeds from the general metamorphosis of the non-nitrogenous and nitrogenous animal matters.

*Uses.*—Since we are as ignorant of the chemical changes which taurocholic acid undergoes in the intestinal canal, as we are regarding those of glycocholic acid, we are unable to express by a chemical equation, the part which it takes in the process of digestion; and until this can be done, we cannot give a satisfactory explanation of the *chemical* action of the bile. The consideration of the *physiological* relations, from which we judge of the importance of the biliary secretion, in reference to the metamorphosis of the animal tissues and to animal life, and which is based on the chemical substratum we have here laid down, will be found in another part of this work.

*Pneumic* (or *pulmonic*) acid probably belongs to this group of conjugated nitrogenous acids. This acid, of which as yet we know very little, was discovered by Verdeil<sup>1</sup> in the tissue of the lungs. The minced pulmonary tissue is stirred with water and exposed to strong pressure; the decanted acid fluid is heated in order to coagulate the albumen, and is then filtered, neutralized with baryta-water, and evaporated to three-fourths of its volume. After the removal of albuminous and some other matters by sulphate of copper, and the excess of the copper by sulphide of barium, we evaporate the fluid till crystals of sulphate of soda are formed; we then add a little sulphuric acid, and boil with alcohol. The acid gradually separates from the alcoholic solution on cooling. It crystallizes in oblique rhombic prisms, is extremely glistening, and refracts light strongly, loses no water of crystallization at 100°, but at a higher temperature decomposes. It dissolves readily in water, is insoluble in cold but dissolves in boiling alcohol, is insoluble in ether, forms crystallizable salts with bases, and contains not only carbon, hydrogen, and oxygen, but also nitrogen and sulphur.

## HALOID BASES AND HALOID SALTS.

The consideration of the above series of organic acids has made us become acquainted with a number of bodies, which, in opposition to the ordinary rules of chemistry, enter into combination with acids without

<sup>1</sup> Compt. rend. T. 83, p. 604, and *Traité de Chimie anatomique et physiologique*, T. 2, p. 460.

depriving them of their most essential chemical characters. There is, however, also a series of substances which can so combine with organic and mineral acids, that they perfectly neutralize their acidity, and can form with them true salts, both neutral and acid, without deserving, on account of their containing no nitrogen, to be classed among the alkaloids.

This class of salts has recently been referred to the conjugated compounds (by Gerhardt and Laurent,<sup>1</sup> and Strecker),<sup>2</sup> since the idea of bodies of this nature has become tolerably firmly established; but the property of these non-nitrogenous bases, perfectly to saturate the strongest mineral and organic acids, appears to us a very stringent reason why these bodies should be separated from the true adjuncts, and why their neutral and acid combinations with acids should be separated from the true conjugated acids. Berzelius<sup>3</sup> has applied the name of *Haloids* to these salt-like combinations of acids with non-nitrogenous bodies. If we attempt to apply the highly probable (but not indubitably established) hypothesis of conjugated ammonia, to explain the basicity of the true nitrogenous alkaloids, we shall find such a mode of explanation perfectly inapplicable to these non-nitrogenous bases. These haloid bases may be classed as analogous bodies to oxide of ammonium. For as, according to the ammonium-theory of Berzelius, we assume, in the so-called ammonia-salts, the existence of the oxide of a combination of nitrogen and hydrogen,  $H_4N$ , in which this, in some degree, simulates a metal, so also we are equally justified in seeking for the basicity of these substances in the oxide of a carbo-hydrogen; and more especially since we are already acquainted with pure carbo-hydrogens possessing decided basic properties, as, for instance, the non-oxygenous ethereal oils. This assumption is not in the least opposed by the circumstance that the carbo-hydrogens, like the ammonium, combine with oxygen to form basic oxides. It is true that such a mode of viewing the subject leads us back to the frequently attacked, but by no means perfectly controverted or exploded theory of organic radicals; but, in a department of science so young as chemistry still is, that is the most satisfactory mode of contemplating the subject, which enables us to represent and explain, in the simplest manner, the largest number of analogous phenomena.

These oxides of the carbo-hydrogen radicals are, however, in their isolated state, so different from the known mineral bases and organic alkaloids, and exhibit such weak basic properties, that for a long period it was altogether denied that they possessed the character of a base. It is with difficulty that they combine either with acids or with water. Even their hydrates differ so greatly from the anhydrous oxides, that they were formerly regarded as perfectly different bodies, and ether was carefully distinguished from alcohol, oxide of amyl from fusel oil, and oxide of methyl from pyroxylic spirit. Moreover, it is only with difficulty, and in certain instances, that we can separate the water from these hydrates. In the same way, their combinations with acids, although most of them are perfectly neutral, bear very little resemblance

<sup>1</sup> Ann. d. Chim. et de Phys. 3 Sér. T. 24, pp. 163-208.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 68, S. 47-55.

<sup>3</sup> Jahresber. 27, S. 425.

in their character to salts, and hence most of them have received trivial names, as, naphthas, fats, &c.

As has been already mentioned, the haloid bases form neutral as well as acid salts; in the former the acidity of the stronger acids is, for the most part, far more perfectly neutralized than in the salts of the nitrogenous alkaloids; for the neutral salts, with a few exceptions, exert no action on litmus; they are, however, essentially distinguished from the salts of almost all other known bases, by the circumstance that they cannot be so readily separated from their acids by simple or double elective affinity. The haloids cannot be decomposed by stronger acids, nor yet by stronger bases; it requires a more considerable time, and a more prolonged action of heat to resolve them into their proximate constituents, than is necessary for ordinary salts.

In these decompositions of the haloid salts we constantly find that the base, during its liberation, combines with water, and is thus separated as a hydrate (for instance, not as oxide of ethyl, but as alcohol; not as oxide of methyl but as pyroxylic spirit, not as oxide of lipyl but as glycerine). Conversely, the haloid bases in uniting with acids give off all their water, so that they always form perfectly anhydrous salts—a fact of which chemists have long availed themselves, in order to ascertain the composition of organic acids in the anhydrous state; (the combinations of such acids with oxide of ethyl or oxide of methyl, being submitted to examination.)

We should fall into a great error, if we were to conclude from the peculiar relations of the haloids that organic bodies are constituted on entirely different principles from mineral bodies; for the chemical laws deduced from pure inorganic compounds meet with their fullest application in these compound organic matters; it is, however, inorganic chemistry which teaches us, that the smaller the chemical attraction between two substances, with so much the more difficulty can they combine with one another, but when once combined, they often resist the most powerful decomposing agents; we need only refer by way of illustration, to the relations of silicic and phosphoric acids to alumina and zirconia. A natural law admits of no exceptions, and if the principles taking their origin in inorganic chemistry be true natural laws, they must be applied, in their fullest extent, to the chemical combinations of organic matters.

The true nature of the acid salts of the haloid bases was also for a long period not recognized; these substances were regarded as peculiar acids, whose consideration led indeed very materially to the theory of conjugated acids and conjugation; but there is an essential difference between an acid haloid salt and a conjugated acid. We have already seen that in the conjugated acids, the true acid has lost none of its saturating capacity, while in these acid haloids half of the acid is always saturated by the haloid base: we know, for instance, that sulphovinic acid cannot, by any possibility, be regarded as a conjugated acid, since only half of the sulphuric acid contained in it, is in a state to saturate a base, just as in bisulphate of potash only half of the acid can be engaged in saturating the base. Notwithstanding this very striking difference, many of the acid haloid salts are, unfortunately, still ranked amongst the conjugated acids.

Moreover, these acid salts are distinguished from the other known acid salts of other bases by the difficulty with which the true base can be separated from the compound; indeed, the separation is here, for the most part, more difficult to accomplish by strong affinities than in the neutral haloid salts. The acid haloids have, however, very many properties in common with one another; they are either solid and crystallizable, or liquid, and, like most of the acid salts in mineral chemistry, always contain 1 atom of water, from which they cannot be separated without total decomposition, except by means of a base; further, however volatile the acid and the base may be, these acid salts cannot be distilled or sublimed undecomposed; and, lastly, it is worthy of remark that their combinations with bases, are almost without exception soluble in water, even though the acid in question formed ever so insoluble a salt with a base (as, for instance, in the case of sulphate of oxide of ethyl, and baryta).

Amongst the haloid bases there is a series of homologous bodies of high interest in relation to theoretical chemistry, but scarcely falling within the sphere of zoo-chemistry. These are the bodies already mentioned in p. 48, possessing the general formula  $C_nH_{n+1}O$ , and standing in a definite relation to the acids of the first group.

There is, however, another haloid base of more importance in zoo-chemistry, but homologous to no other body with which we are acquainted, the *oxide of lipyl*, which, in combination with the fatty acids, constitutes the fats which hold so prominent a place in physiological chemistry. There are many other haloid bases, but for the most part only some of their combinations, namely, their acid salts, have been examined; and in their isolated as well as in their hydrated state they are yet unknown. Hence, we have here only to consider *oxide of lipyl* and its combinations, and *oxide of cetyl*, which is homologous to the group of ethers.

#### OXIDE OF LIPYL.— $C_3H_2O$ .

On boiling one of the common fats or fatty oils with a caustic alkali, with the hydrate of an alkaline earth, with hydrate of magnesia, or oxide of zinc or of lead, the fat, without assimilating oxygen, or giving off hydrogen, is decomposed into one or more *fatty acids*, which combine with the base that has been employed, and form *soaps*, and a peculiar sweet matter, *glycerine*. On comparing the weight of the resulting products of decomposition with that of the fat which was employed, we find that an increase of weight has taken place in consequence of an assimilation of water.

In order to explain the nature of this process, it was assumed that the fats are combinations similar to the salts of oxide of ethyl, and that *glycerine*, represented by the formula  $C_3H_2O$ , constituted the base of the fats; but the constitution of glycerio-sulphuric acid proves that *glycerine* must be represented by the formula  $C_6H_7O_5$ , and that consequently it cannot be regarded as the base of the neutral fats. Hence it is probable that the fats contain, in addition to the fatty acid, the oxide of a radical, having the composition which was formerly ascribed to *glycerine*; and that this oxide in its separation from the fatty acid assimilates



water, and is converted into another body, as in the case of oxide of ethyl when it is expelled by an acid from its combination. To this hypothetical radical, Berzelius has applied the name of *lipyl*.

That the base in the fats is not glycerine seems obvious also from the circumstance that hitherto no neutral fat has been prepared from glycerine and the fatty acids. Whether the butyrin that has been artificially formed from glycerine and butyric acid has the same composition with that contained in butter has not yet been ascertained. Acrolein, which is polymeric with oxide of lipyl, and is a product of distillation of glycerine, cannot, any more than glycerine, be the base of the fats, since it cannot be made to combine even with strong acids.

This conversion of the fats into acids and glycerine, may be induced by other bases than those we have already mentioned, namely, by the soluble carbonates and borates, if they be digested with the fats for a sufficiently long period.

In the case of the carbonates we must, however, suppose that in this process the alkaline carbonate is first resolved into alkaline bicarbonate and free alkali, and that it is the latter only which takes part in the saponification; and that, on further boiling, the alkaline bicarbonate loses 1 atom of carbonic acid, and becomes converted into a simple salt, which again acts on the fat in the above-described manner.

Ammonia and its carbonate only form soaps after a more prolonged action.

### GLYCERINE.— $C_6H_7O_5.HO$ .

#### *Chemical Relations.*

*Properties.*—Glycerine is a faintly yellow fluid with an agreeable, sweet taste; it attracts water from the atmosphere, dissolves readily in water and alcohol, but not in ether, and exerts no reaction on vegetable colors. It dissolves alkalies and several of the metallic oxides (for instance, oxide of lead) in large quantities; in a concentrated state, it admits of being distilled with only partial decomposition, but when rapidly heated, it is entirely decomposed; if its watery solution be exposed to evaporation, decomposition immediately commences: when heated in the air, it becomes inflammable, and burns with a blue flame. If heated with anhydrous phosphorus in a tube from which fresh air is excluded, it yields acrolein. If glycerine be dissolved in a large quantity of water, mixed with yeast, and exposed to a temperature of between  $20^\circ$  and  $30^\circ$ , it develops a small quantity of gas, and is converted into metacetic acid ( $C_5H_7O_5 - 2HO = C_5H_5O_3$ ; Redtenbacher).<sup>1</sup> Treated with spongy platinum, glycerine also becomes converted into an acid (Döbereiner).<sup>2</sup> By concentrated nitric acid it is converted into carbonic acid, oxalic acid, and water; with hydrochloric acid and peroxide of manganese, it yields a large quantity of formic acid.

*Composition.*—In accordance with the above formula deduced by Pelouze<sup>3</sup> from his analyses of pure glycerine and its acid salts, this substance consists of:

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 174–177.

<sup>2</sup> Journ. f. pr. Ch. Bd. 29, S. 451.

<sup>3</sup> Compt. rend. T. 21, pp. 718–722.

Carbon,	.	.	.	.	.	6 atoms,	.	.	.	.	39.130
Hydrogen,	.	.	.	.	.	7 "	.	.	.	.	7.609
Oxygen,	.	.	.	.	.	5 "	.	.	.	.	43.478
Water,	.	.	.	.	.	1 "	.	.	.	.	9.783
											<hr/> 100 000

The atomic weight of anhydrous glycerine = 1037.5.

Glycerine cannot be regarded as a hydrate of oxide of lipyl, because in its combinations it always contains 3 atoms of water more than a double atom of oxide of lipyl; and we know that no haloid base retains its hydrate-water when it combines with acids.

*Combinations.*—No neutral salts of glycerine have yet been exhibited, but we are acquainted with several of its acid salts, which, like the acid salts of the oxides of ethyl and methyl, unite with bases, and form a series of compounds.

*Bisulphate of glycerine (glycero-sulphuric acid)*  $C_6H_7O_5 \cdot SO_3 + HO \cdot SO_3$ , is formed by the direct union of glycerine with sulphuric acid; the excess of sulphuric acid is removed by saturating with carbonate of lime or baryta; the sulphate of glycerine-lime or glycerine-baryta is decomposed with oxalic acid and the filtered fluid evaporated *in vacuo*.

This acid salt forms a colorless fluid, which, on evaporation even *in vacuo*, is readily decomposed into glycerine and sulphuric acid; it has a strongly acid taste, reddens litmus, and forms easily soluble double salts, even with baryta and lime. These salts readily yield glycerine when boiled, and even more readily when treated with an excess of base; the dry salts when heated carbonize and develop a vapor (containing acrolein) with an extremely disagreeable odor, and irritating to the eyes. The lime-salt crystallizes in colorless needles, and  $= CaO \cdot SO_3 + C_6H_7O_5 \cdot SO_3$ .

*Acid phosphate of glycerine (glycero-phosphoric acid)*,  $C_6H_7O_5 \cdot 2HO + PO_5$ , is obtained by the direct action of syrupy glycerine on pulverized glacial phosphoric acid, which develops much heat, the temperature even rising to  $100^\circ$ . The excess of phosphoric acid is removed by baryta, and the baryta-salt decomposed by sulphuric acid. When in a concentrated state the body in question forms a colorless fluid, which even *in vacuo* cannot be very strongly concentrated without undergoing decomposition; it does not crystallize, has a strongly acid taste, and dissolves freely in water and alcohol; with bases it forms double salts, which dissolve readily in water, but so very slightly in alcohol that this fluid precipitates them from their aqueous solutions. *Phosphate of glycerine-lime*,  $2CaO + C_6H_7O_5 + PO_5$ , crystallizes in white, glistening scales, and dissolves in cold water; it is, however, so slightly soluble in hot water that it is precipitated from its aqueous solution by boiling. The baryta-salt contains 1 atom of tribasic phosphoric acid, 2 atoms of baryta, and 1 atom of glycerine.

*Bitartrate of glycerine*,  $C_6H_7O_5 \cdot C_4H_2O_5 + HO \cdot C_4H_2O_5$ , is produced, according to Berzelius,<sup>1</sup> on heating 1 part of glycerine, dried at  $120^\circ$ , with 2 parts of dry tartaric acid; it is a semi-solid transparent body, which is solid at  $0^\circ$ , but at  $25^\circ$  admits of being drawn out in long threads; it deliquesces in the air, does not dissolve in alcohol, and with

<sup>1</sup> Jahresber. Bd. 27, S. 438.

bases forms soluble uncrystallizable double salts, which are readily decomposed by an excess of base. The relations of *biracemate of glycerine* are similar to those of this salt.

*Products of its metamorphosis.*—*Acrolein*,  $C_6H_4O_2$ , discovered by Redtenbacher,<sup>1</sup> is obtained from glycerine by submitting it to dry distillation with a little anhydrous phosphoric acid in a stream of dry carbonic acid gas; the distillate, consisting of a thick oil, of an acid fluid swimming on it, and of acrolein floating on the latter, must be digested with oxide of lead and distilled at  $52^\circ$  into a receiver containing carbonic acid, by which means we obtain the acrolein. It is an oily fluid, which strongly refracts light, has an acrid, burning taste, irritates the eyes and respiratory organs, and forms a neutral solution in water devoid of air, which, however, very soon assumes an acid reaction on exposure to the atmosphere. It instantly reduces oxide of silver, and it decrepitates both with nitric acid and with potash.

*Acrylic acid*,  $C_6H_3O_3 + HO$ , is formed when acrolein is oxidized either by exposure to the air or by oxide of silver; it is a limpid fluid, with an odor resembling that of very strong acetic acid, and a pure, acid taste; nitric acid converts it into acetic and formic acids; it forms soluble, crystallizable salts with bases.

*Disacrone, disacryl*,  $C_{10}H_7O_4$ , is gradually deposited from acrolein exposed to the atmosphere; it is idio-electric, devoid of odor and taste, and insoluble in all *menstrua*.

*Preparation.*—Glycerine is formed, as we have already mentioned, during the saponification of the fats, from the oxide of lipyl contained in them combining with 4 atoms of water. It is usually prepared from the aqueous fluid which separates during the preparation of lead-plaster, and contains it, together with oxide of lead, in solution. After the removal of the lead by sulphuretted hydrogen we concentrate the solution first in the water-bath and subsequently *in vacuo*. We may also obtain it from the mother-liquid yielded in ordinary saponification by the alkalies, on saturating the alkali of the lye with sulphuric acid, then heating it with carbonate of baryta, evaporating the filtered fluid, and extracting with alcohol. It may be obtained very readily, and in a state of purity, by dissolving castor-oil in absolute alcohol, and passing hydrochloric acid gas through the fluid; at the end of the operation the compounds of the fatty acids with oxide of ethyl, which have been produced, must be separated by means of water. The aqueous fluid, on evaporation, leaves glycerine, which may be entirely freed from adhering traces of the fatty ethers by being shaken in ether.

*Tests.*—Glycerine could not be readily detected in animal fluids unless we were able to obtain it in sufficient quantity to admit of its being subjected to an elementary analysis; but this would be hardly possible, since it would be difficult to obtain the glycerine in a state of purity from the animal fluids. Fortunately, however, acrolein is a substance with so intense and characteristic an odor that this product of the decomposition of glycerine may be employed as a test of its presence. The glycerine, separated in as pure a state as possible, must be rapidly heated either alone or with a little anhydrous phosphoric acid, when, if

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 47, S. 113-148.

the glycerine be much diluted, the peculiar and very disagreeable odor, not unlike that developed by the wick of an expiring oil-lamp, is evolved with sufficient distinctness.

### *Physiological Relations.*

*Occurrence.*—Glycerine has been recently discovered by Gobley<sup>1</sup> in animal bodies. He first detected it in the *yolk of the egg* of the common fowl in the form of phosphate of glycerine-ammonia, and subsequently<sup>2</sup> in the same state of combination in the *fats of the brain*.

*Origin.*—Regarding the source of the glycerine in the organism, there can be no doubt that, in addition to the true fats—the stearate, margarate, and oleate of oxide of lipyl—there are many fatty acids, either free or in combination with alkalies, occurring in the animal body. Since the combinations of the fatty acids and oxide of lipyl are introduced into the animal body from without, we need not wonder that glycerine, which is formed from oxide of lipyl during the decomposition of the fats, is not found in far larger quantity in this or that animal fluid. We have already directed attention to the possibility (p. 62 and p. 106) that in the consumption and gradual oxidation of the neutral fats, the oxide of lipyl, separated as glycerine, is probably converted into lactic or even into metacetic acid. Further investigations are, however, necessary before we can decide whether this conjecture is of any real value. The uses of fatty articles of food would thus assume a new aspect, since they would in this way contribute to the formation of the free acids which act so important a part in many of the processes of animal chemistry.

How the glycerine in the yolk of egg and in the brain becomes associated with the phosphoric acid, we cannot specially explain, but, considering the frequency with which phosphorus occurs, both in its unoxidized state and as phosphoric acid, there is nothing singular or inexplicable in such a combination.

### SALTS OF OXIDE OF LIPYL.—FATS.

#### *Chemical Relations.*

*General Properties.*—It is especially worthy of remark that the properties of these haloids are almost entirely influenced by the acids contained in them; while in the salts of oxide of ethyl, most of the properties, including those of the most general character, appear to depend principally on the base, and to be altogether independent of the nature of the acid. Hence we find the properties of the neutral fats to be extremely similar to those of the fatty acids already described (from p. 103 to p. 112).

Most of the animal fats are soft and greasy at an ordinary temperature, although some are firm and waxy, and a few liquid; they almost all correspond, however, in the following points. When exposed to strong cold, especially when in solution in alcohol, they may be obtained

<sup>1</sup> Compt. rend. T. 21, pp. 766-769, et 988-992.

<sup>2</sup> Journ. de Pharm. 3 Sér. T. 11, pp. 409-417, et T. 12, pp. 5-13.

in white scales or minute plates of a peculiar lustre; when perfectly pure, they are for the most part colorless and transparent, they swim on water, render paper and linen transparent, are bad conductors of electricity and heat, melt for the most part below the boiling-point of water, are altogether decomposed when distilled, unless the process be conducted *in vacuo*, and are devoid of smell and taste when they are pure and fresh; they are insoluble in water, but most of them dissolve in boiling alcohol, from which they again separate on cooling; they are all soluble in ether and in volatile oils; when perfectly pure they exert no reaction on vegetable colors, but on exposure to the air many of them readily become rancid and acid from the absorption of large quantities of oxygen. When exposed to a strong heat, and free access of oxygen is admitted, they are inflammable, and burn with a clear flame.

There are certain ferments which resolve the fats into glycerine and the corresponding fatty acid, in the same manner as sugar is resolved into alcohol and carbonic acid, or salicin into saligenin and sugar, or amygdalin into sugar, hydrocyanic acid, and oil of bitter almonds. Albuminous substances which have already undergone a certain degree of decomposition (putrefaction) act in this manner as ferments to the fats.

If we mix putrid fibrin, which forms an albuminous fluid, with water, or putrid casein with fat, so as to form an emulsion, and digest the mixture for some time at a temperature of 37°, the corresponding fatty acids separate from the oxide of lipyl, which very soon undergoes further alterations. In the fermentation of milk, where sugar is present, it appears from my investigations<sup>1</sup> that the fats are decomposed in precisely the same manner as if merely the putrefying protein-compounds were acting as ferments, and as if no sugar were present. Cl. Bernard<sup>2</sup> on digesting fats with pancreatic fluid observed that they were decomposed into fatty acids and glycerine, from which he concluded that during the act of digestion the fats are constantly decomposed into glycerine and fatty acids—a conclusion, however, still admitting of considerable doubt.

By *dry distillation* certain fats yield other fatty and inflammable substances, and leave a little charcoal; others are in part converted into peculiar fatty acids. When very rapidly heated or thrown on incandescent bodies, they carbonize and develop olefiant gas.

The fats are decomposed by prolonged contact with *chlorine*, *bromine*, and *iodine*; while, on the other hand, they take up sulphur, selenium, and phosphorus, without undergoing any change; with the former, they only undergo decomposition on the application of heat.

By concentrated *mineral acids* they are for the most part converted into fatty acids, and on the application of sulphuric acid, they yield acid sulphate of glycerine.

*Stearate of oxide of lipyl*, *stearin*, occurs as a pure white substance; it separates on cooling from its alcoholic solution in snow-white, glistening scales; under the microscope it appears chiefly in the form of quadrangular tablets, which, although almost square, are, according to

<sup>1</sup> Simon's Beitr. Bd. 1, S. 63–76.

<sup>2</sup> Arch. génér. de méd. 4 Sér. T. 19, p. 73.

Schmidt,<sup>1</sup> rhombs with angles  $\approx 90^\circ 5'$ , but sometimes in the form of short rhombic prisms (thick rhombic plates), whose surfaces, according to Schmidt, are inclined to one another at angles of  $67^\circ 40'$  and  $52^\circ 40'$ . It melts at  $+62^\circ$ , solidifies, but does not become crystalline on cooling, is brittle, when dry is not a conductor of galvanic electricity, is insoluble in cold and only slightly soluble in hot alcohol, but dissolves very readily in ether. On dry distillation it yields stearic and margaric acids, and the products of decomposition of glycerine; on saponification it yields stearic acid and glycerine.

*Margarate of oxide of lipyl, margarin*, is white and solid; it crystallizes from alcohol as a flocculent white powder, which under the microscope appears in the form of very delicate and often curved needles, which are so grouped as to radiate from one point as a nucleus, and thus to form a whorl of fine, capillary threads; it melts at  $+48^\circ$ , and dissolves slightly in alcohol but readily in hot ether; it separates from either solution on cooling in nacreous scales, and on saponification yields glycerine and margaric acid.

*Oleate of oxide of lipyl, olein, or elain*, is a colorless oil which solidifies at a low temperature, is not a conductor of galvanic electricity, becomes rancid on exposure to the air, is never entirely free from margarin and stearin, and on saponification yields, in addition to glycerine and oleic acid, a much larger quantity of margaric acid than can be supposed to be derived from the decomposition of the margarin.

*Preparation.*—The above fats may be obtained in various ways, although seldom in a state of perfect purity, from the fat contained in cellular tissue, by repeated melting and purification with water. Usually we dissolve the fat in boiling alcohol, from which, on cooling, the stearin, and a great part of the margarin, separate in crystalline scales, while the olein is almost the only substance remaining dissolved in the cold alcohol. Margarin is obtained in the greatest purity from the hot alcoholic solution of those fats, which, like human fat and the vegetable fats, contain no stearin; moreover, by strong pressure between the folds of filtering paper, the olein may be tolerably effectually separated from the stearin and margarin, since, above a certain temperature, it penetrates the paper. Tolerably pure olein may be obtained by digesting a fat with half the quantity of potash required for its complete saponification; in this case the stearin and margarin are saponified, while the olein remains unchanged. The corresponding acids may be obtained in a similar way, but in a state of much greater purity.

*Tests.*—Cases sometimes present themselves in which it is not easy to ascertain whether the substance to be examined contains salts of oxide of lipyl, or the corresponding fatty acids. In dealing with small quantities, we obviously cannot rely on the acid reaction, or on the formation of glycerine; in such cases the simplest method is to obtain an ethereal extract of the alcoholic extract to which a little acetic acid had been added, and then, by digestion with water, to separate the residue of the ethereal solution from other substances. The remaining fat is then to be dissolved in alcohol, and to be treated with an alcoholic solution of acetate of lead. If the addition of ammonia give rise to no precipitate,

<sup>1</sup> Entwurf u. s. w. S. 84.

it is a proof that the solution contains no free fatty acids, but only salts of oxide of lipyl.

Free fat in the animal fluids, tissues, and cells, is most commonly and, indeed, most satisfactorily detected by the microscope; the vesicles in which fat ordinarily appears, present so characteristic an appearance, that when they have been seen for a few times under the microscope, they can hardly be confounded with anything else; the more consistent fat, containing little olein, sometimes, however, occurs in nodular, sausage-shaped, and only faintly-transparent clumps, which cannot so readily be recognized as fat. In these cases, chemistry must come to the aid of microscopic investigation, as, for instance, where the fat-vesicles in cells are so minute, that, with the highest magnifying powers, they appear as mere dark points or granules. Many histologists now maintain that these points and aggregate granules may be very readily distinguished under the microscope, by their solubility in ether; but the extraction of the fat from the cells by ether, is by no means easy, for its rapid evaporation under the microscope, renders it very difficult, if not impossible, to observe the individual cells. Before making our observations we must, therefore, repeatedly pour a little ether on the object, and allow it again to run off, or if we have fine sections of tissue, we may digest them in ether. Unfortunately, however, the cells and other histological elements are often so distorted by ether, that even after long maceration in water, an accurate observation is no longer possible; and it is nearly the same in most cases with alcohol, by which, however, well-prepared sections of many parts, as, for instance, nerve-fibres, may often have their fat thoroughly removed. Moreover, alkalies cannot be advantageously applied to the partial saponification of these fats, since they often dissolve albuminous parts much sooner than the fats. We shall see, in a future part of this work, that some histologists believe that they have found fat-granules in tissues which have been hitherto regarded as utterly devoid of fat; and have been too hastily led, by imperfect experiments, to form theories regarding the fatty degeneration of cells and tissues.

### *Physiological Relations.*

*Occurrence.*—Fats occur, not only in the animal world, but also in vegetables, especially in seeds and the kernels of fruits, from which we chiefly obtain the fatty oils and certain butter-like fats, as for instance, cacao butter, palm oil, &c. Fats have been found in almost all parts of all animals, and it is only in the lowest classes of animals, that fat is entirely absent. It is in the higher organisms that we find most fat, where it exists as a mixture of the above-named salts of oxide of lipyl, and is deposited in the cellular tissue in oval or polyhedric cells.

It is very rarely that we find one of the above-named fats unmixed with the others, and in the few cases of this nature which have been observed, the character of the fat has been recognized by the microscope only, and not by chemical means; thus C. Schmidt (according to Bergmann<sup>1</sup>) and Vogt<sup>2</sup> found distinct crystals of stearin in the ovum of

<sup>1</sup> Müller's Arch. 1841. S. 89.

<sup>2</sup> Entwicklung der Geburtshelferkröte. Solothurn. 1842. Einl.

the frog, and of the accoucheur toad (*Bufo obstetricans*), and I have frequently, although not invariably, found delicate masses of acicular crystals in the albumen of eggs that had been sat upon from three to six days, which from the few tests that I could attempt, seemed to consist of margarin.

When we consider the occurrence of fat in the different parts of the human body in a normal condition, we, in the first place, discover large accumulations of fat, which, when constituting an integral constituent of certain organs, rarely disappear entirely, even in the latest stages of wasting diseases; in the next place we observe, that there are parts of the body in which the quantity of fat varies considerably, being either extraordinarily small or very large; and finally, that there are some organs in which accumulations of fat are of very rare occurrence. The orbit of the eye and the heart appear to be the most-constant seats of fat, for although we observe that the fatty matters surrounding the different parts of the eye diminish in all forms of disease, causing the eyeball to sink in the orbit, the socket of the eye is never found entirely free from fat. A similar remark applies to the fat surrounding the heart, and penetrating between its bundles of fibres; and it would likewise appear that fat is never entirely absent from the muscles of the face, for every one who has dissected these muscles must have noticed how largely the human face is furnished with fat.

Large quantities of fat, not constituting so essential and integral a part of the organs, and often almost entirely disappearing, are principally found under the cutis and in the cellular tissue, investing the muscles; in the interstices of several of the larger muscles, about the glutæ, on the soles of the feet, and in the inner surface of the hands. Fat is frequently found deposited in sacs round different tendons projecting between the ends of the bones into the joints, where they form special accumulations of fat, known by the name of the *Haversian glands*. Large deposits of fat are generally found in the omentum, and surrounding the kidneys, constituting the *folliculus adiposus renum*, which usually contains a harder fat, having a larger quantity of margarin, than occurs in other parts of the body.

The female breast is always so largely interspersed with masses of fat, that full prominent breasts frequently yield a small quantity of milk, being enlarged solely by the deposition of fat.

The marrow of the bones consists, for the most part, of fat, which not only remains undiminished, but is even not unfrequently largely augmented in various diseases of the bones, as, for instance, in *osteomalacia*. This bone-fat is perfectly identical with the ordinary fat of the cellular tissue, excepting that it contains somewhat more olein, especially where there is *osteomalacia*.

All other parts of the animal and more especially of the human body, are penetrated by fat. The smallest quantity, and indeed, occasionally, not a trace of fat is to be found in the pulmonary tissue, in the glans penis and the clitoris, and, if we except the so-called non-saponifiable fats, in the brain.

Many organs have a special tendency to accumulate large quantities of fat, when in pathological states; hence it is especially necessary to



determine the normal quantities of fat which these organs contain. We particularly refer to the *liver*, *spleen*, and *kidneys*. We do not refer to those special fat-cells surrounded by connective tissue, such as we find in the *Follicules adiposus cutis et renum*; but we here find the fat specially accumulated in cells not very unlike the ordinary epithelial cells. On making a microscopic examination of the liver in its perfectly normal condition at a certain period after a meal, it is very rarely that we find the hepatic cells perfectly free from fat. In the spleen, which naturally contains so large a number of colorless cells, we always find fat both in the carnivora and herbivora. Frerichs<sup>1</sup> discovered fat in the perfectly normal kidneys of dogs and cats, and von Hessling<sup>2</sup> constantly found it in the kidneys of fishes; and, finally, Lang<sup>3</sup> has demonstrated its ordinary occurrence in the kidneys of cats by microscopical and chemical investigations. In normal human kidneys, Frerichs observed small quantities of fat, and Lang found that fat was as often present as absent. Lang found from 1.8% to 3.9% of fat in the dried substance of the kidneys in cats, but he was unable to detect the presence of this substance in the kidneys of an ox or of a calf. It appears, both from pathological observations as well as from these investigations of normal kidneys, that the fat principally occurs in the cortical substance, and that it exists in the form of minute drops, which are partly free and partly enclosed in the cells of the tubules.

I must not omit to mention an observation which I have repeatedly made. I have seen the "canaliculi contorti" of the kidneys of three freshly-killed deer, and of several hares, filled with free fat-globules, and the epithelium similarly filled, while scarcely an isolated fat-globule could be seen in the true tubular substance or in the contents of the bladder of these animals. Since these animals were perfectly healthy, and had certainly taken no fatty food shortly before their death, these observations to a certain degree oppose Lang's view, according to which the amount of fat in the normal kidneys is dependent upon the use of fatty food. At all events the subject requires further investigation, notwithstanding Lang's careful observations. On a recent occasion I could find no fat in the kidneys of a deer: could those three animals have been in a special state of development?

[We may here notice the investigations of Professor Beale<sup>4</sup> in reference to the amount of fat in the human liver and kidney both in health and disease.

Beale gives the results of his analyses of one kidney in a state of fatty degeneration, of four diabetic kidneys, and of two diabetic livers.

Taking the amount of fat in 100 parts of the *solid matter*, it was found that one diabetic kidney contained five times the normal quantity of fat; another four times as much; a third nearly the same proportion; and the fourth contained three times as much as was found in the healthy kidney. The fatty kidney contained rather more than six times the quantity of fat (26.97%) obtained in the healthy specimen.

<sup>1</sup> Die Bright'sche Nierenkrankheit u. deren Behandlung. Braunschweig, 1851, S. 43.

<sup>2</sup> Histol. Beitr. z. Lehre v. d. Harnabsonderung. Jena, 1851, S. 52.

<sup>3</sup> De adipe in urina et renibus, diss. inaug. Dorpat, 1852, p. 48-64.

<sup>4</sup> British and Foreign Medico-Chirurgical Review, Vol. 12, p. 226.

Of the two diabetic livers, one contained only 4·64, and the other 7·85% of fat; while in the two healthy livers the corresponding numbers were 12·15 and 15·81%.

HEALTHY LIVER.				
	I.	100 parts of solid matter.	II.	100 parts of solid matter.
Water, . . . . .	68·58	...	72·05	...
Solid matter, . . . . .	31·42	...	27·95	...
Fatty matter, . . . . .	3·82	12·15	4·28	15·31
Extractive soluble in water and alcohol, . . . . .	10·07	32·04	10·40	37·20
Extractive soluble in water only, and albumen, . . . . .				
Alkaline and earthy salts, . . . . .	1·50	4·77	1·19	4·20
Matter insoluble in water, alcohol, and ether, . . . . .	16·03	51·01	12·08	43·22

HEALTHY KIDNEY.		
	I.	100 parts of solid matter.
Water, . . . . .	76·450	.....
Solid matter, . . . . .	23·550	.....
Fatty matter containing much cholesterin, . . . . .	·939	3·98
Extractive matter soluble in water, . . . . .	5·840	24·79
Fixed alkaline salts, . . . . .	1·010	4·28
Earthy salts, . . . . .	·396	1·68
Albumen, vessels, &c. . . . .	15·865	65·24

It would thus appear that in diabetes the kidney, in its chemical composition, approaches to that of fatty degeneration, while the liver appears starved, and its secreting cells seem to manifest a tendency opposite to that of fatty degeneration.—G. E. D.]

We have already spoken of the occurrence of fat in the *animal fluids*. The amount of fat in the *blood* does not vary much in a normal condition, and is, according to Boussingault's numerous investigations,<sup>1</sup> wholly independent of the amount of fat contained in the food. The blood contains from 0·14 to 0·33% of fat in a normal condition. Boussingault found from 0·2 to 3·0% of fat in the blood of dogs, whether they had partaken of food deficient or abounding in fat, and 0·4% in that of birds. Tiedemann and Gmelin always found the *chyle* very rich in fat; and its milky turbidity, as well as that of the *lymph*, is owing to the fats which it holds in suspension.

<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. T. 24, p. 460.

I was unable to discover any trace of Boudet's *serolin* in the chyle of a dog. The fat which was extracted with ether was oily, was not precipitated from boiling alcohol on cooling, and was for the most part saponifiable.

This seems to confirm Schultz's observation,<sup>1</sup> that the fat of the blood is more consistent than that of the chyle, and it may further be remarked that the fats of the blood are mostly saponified or incapable of saponification, while those of the chyle correspond to the ordinary salts of oxide of lipyl.

The excellent investigations recently instituted by Cl. Bernard<sup>2</sup> have afforded the most striking proof that the fats are digested by the pancreatic fluid; *i. e.*, that the fats are not reduced to an emulsive state, either by the gastric juice, or (as Brodie<sup>3</sup> believed that he had found) by the bile, and thus fitted for resorption. But the conclusion which Bernard would draw from an experiment in which he found that fat had been converted into fatty acids and glycerine by the action of the pancreatic juice, *viz.*, that all fats are converted by the process of digestion into glycerine and the corresponding fatty acids, is controverted by the fact above referred to, that the chyle contains, in comparison with the blood, much unsaponified and but little saponified fat.

Marchand and Colberg found oily and crystalline fat in the lymph.

The quantity of fat in the human body varies considerably at *different periods of life*. Thus in the fœtus we generally find no fat, except a few small masses in the omentum and in the loins. Infants prematurely born are rounder in form immediately after birth than at a subsequent period, for as their organism is not fully prepared for an atmospheric life, they soon become emaciated, and lose much fat through the intestinal canal. The muscular tissues of the heart and face are found to be copiously furnished with fat even at this early period. New-born children are in general tolerably plump and roundish, and have a considerable quantity of fat deposited under the skin. The organism is most rich in fat during childhood, but this deposition of adipose matter diminishes with the development of the sexual functions, although it again increases at a more mature period of life, and then occasionally acquires an excess never observed at any other age. Extreme old age gradually arrests this tendency to adiposity until it is completely destroyed by *marasmus senilis*.

A merely superficial comparison of the *sexes* shows that the female organism contains more fat, and has a greater tendency to the deposition of fatty matter than the male, as indeed is most evident from the rounded outlines and symmetrical curves of the female figure, which cannot be entirely destroyed even by influences most inimical to the deposition of fat.

We find that special physiological relations give rise in some cases to an increase, and in others to a diminution of the fat in the animal organism. Thus an excessive *activity of the sexual functions* prevents the increase of fat, and even induces considerable emaciation where the

<sup>1</sup> System der Circulation, 1836, S. 181.

<sup>2</sup> Arch. Génér. de Méd. 4 Sér. T. 19, pp. 60-81.

<sup>3</sup> Quart. Journ. of Science. Jan. 1828.

sexual activity is of a morbid character. Men and animals that have been castrated, are, on the contrary, much disposed to become fat, as are also women who have ceased to conceive. Many male animals, according to Haller, lose the marrow from their bones in the season of heat.

It is well known that great *muscular activity* not only impedes, but even utterly arrests the deposition of fat. Thus the flesh of the Arabs, and that of all nations living in a state of nature, as well as of most wild animals, contains a very small quantity of fat, while civilized nations and the domestic animals reared for purposes of food are, in general, much fatter, owing to their inconsiderable muscular activity. Most persons are familiar with the fact that horses become much leaner in summer even when better fed, and that they soon grow fat in the winter. The whole art required in fattening domestic animals consists in suffering them to have little exercise and good feeding.

We have daily opportunities of noticing the influence exercised by *food* alone on the deposition of fat; and the degree to which the *temperament* and *conditions of the mind* affect the corpulency or meagreness of the human body is too obvious to require further notice here.<sup>1</sup>

Every physician is familiar with the marvellous rapidity with which fat disappears from the animal body in acute as well as in chronic *diseases*, and we would here only refer to the fact which undoubtedly is well known to many physicians, that tuberculosis very frequently induces very little or no emaciation, even where the pulmonary tissue is already in a great measure destroyed, if the disease be accompanied with certain forms of hepatic disease, as fatty or nutmeg liver. The emaciation is so inconsiderable in these cases, that any one not acquainted with the physical diagnosis of the disease, would be completely deceived as to its character and the amount of danger.

It appears scarcely necessary to remark that *milk* contains a larger quantity of fat than any other animal fluid. An average of 2.9% of fat has been found in woman's milk. This subject we shall however consider more fully in the second volume of this work, when we purpose treating of the increase and diminution of the fat contained in the milk of different animals under different physiological and pathological relations.

Since Güterbock's observations, attention has been directed to the quantity of fat contained in *pus*, which has frequently been found to amount to 5%.

As we have already remarked, the fat in the *blood* is mostly in a state of saponification; but in many diseases, the blood has been observed to contain large quantities of unsaponified fat. Since we purpose entering more fully into this subject when we proceed to the consideration of the morbid conditions of the blood, we will here only observe, that although as is generally supposed, the blood of drunkards frequently presents large accumulations of free fat, this only occurs where there is already some hepatic disease, as for instance, granular liver, whether this be a mere secretion of colloid-like exudation accompanied with decrease of size in the liver, or that species of granular disease in which some of the hepatic lobules present scattered cells infiltrated with fat.

<sup>1</sup> We may refer to the first volume of Haller's *Elementa Physiologiae* for the most copious accumulation of facts bearing on this subject.

*Pathological depositions of fat*, either free or enclosed in cells, occur most frequently in the *liver*, but also in the *kidneys*, the *spleen*, in paralyzed *muscles*, in the *heart*, and other organs, and occasionally (enclosed in a capsule) in *encysted tumors*. This fatty metamorphosis (as it is termed) of some of the organs, will be specially considered in the second volume of this work, in our remarks on the individual tissues and organs. It will be sufficient at present to remark that these so-called fatty degenerations of organs occur either without any previous exudation by the direct deposition of fat in the tissues, the cells, or the areolar tissue, or (as indeed is more frequently the case), after the resorption of the physiological or pathological tissues or exudations, are deposited in their place. The latter case occurs in paralysis of muscles, where they have undergone fatty degeneration, and in osteoporosis and osteomalacia, where the bones, rendered porous by the resorption of their mineral and organic parts, are found, as it were, swimming in fat; a similar process may occur in the fatty degeneration of the spleen and the kidneys, which many have attempted to explain as the third stage, or indeed, as the essential character of Bright's disease. The endeavor to explain such pathological processes by a perfect metamorphosis of albuminous and fibrinous exudations into fat (that is to say, by a direct metamorphosis of the protein-compounds into fat), is purely chimerical and unsupported by the slightest proof.

It is further an undoubted fact that in many cells, whether they be constituents of physiological tissues, or products of pathological exudations, fat occurs accumulated in large quantities, appearing under the form of vesicles, or more frequently of granules, as in the hepatic cells, in the granular cells in old apoplectic cysts, and in the analogous cells in the expectoration in confirmed chronic catarrh; but it is incorrect to suppose that all strongly tinged, punctuated granular cells, contain much fat: we will, however, postpone all further consideration of this subject to the second volume.

We have no accurate observations regarding the quantity of fat contained in the *fæces* in different diseases; and I will here only remark, that I have always found fat in the normal excrements, but more especially in the stools in diarrhoea; in most of the cases, in which observations have been made regarding an excess of fat in the *fæces*, we are unable to determine whether its increase be owing to the food, or to fatty medicines.

A firm margarin-like fat, has been frequently noticed as present in the excrements of diabetic patients (Simon,<sup>1</sup> Heinrich<sup>2</sup>), but I have never observed any decided increase in the quantity of fat in the *fæces* in diabetes: and the discharge of fat by the intestines cannot therefore be regarded as a constant symptom.

It is equally difficult to form a correct opinion of the quantity of fat in the *urine*. No reliance is to be placed on the older observations, since the presence of fat in the urine was at that period often diagnosed, whenever, in consequence of an alkaline reaction, the urine was covered with a pellicle; this was regarded as fat, although consisting in reality of nothing more than earthy matters. Where the microscope shows fat-

<sup>1</sup> Beitr. Bd. 1 S. 408.

<sup>2</sup> Hæser's Arch. Bd. 6, S. 306.

globules in the urine, they frequently, in women, arise from the external genitals. It is only in slow fevers that I have been able to confirm the old view, and often, but not invariably, to detect fat-globules. In the urine of pregnant women, which contains the so-called kysteine, I have, however, always observed a soft buttery fat. I have never met with true milky, or chylous urine, where the turbidity and color were owing to the presence of fat; for this species of urine seemed to owe its peculiar character to a large quantity of pus-corpuscles, held in suspension, which in all the cases I examined, originated in the kidneys, and were not dependent on vesical catarrh. Where milky urine has been found to contain a large quantity of fat, it may be owing, as in Rayer's case,<sup>2</sup> to milk that had been purposely added, in order to deceive the physician.

It would be very important, in reference to the diagnosis of Bright's disease, if we could confirm the conjecture advanced by Oppolzer, that in this disease, at any rate when there is fatty degeneration of the kidneys, the urine contains fat. I have, unfortunately, hitherto been unable to confirm this conjecture, for even where a *post-mortem* examination showed decided fatty degeneration of the kidneys, the urine exhibited no microscopic fat-globules, nor did the ether extract any trace of fat. In one case only, where the urine removed from the bladder after death, contained the well-known epithelium cylinders, could I discover fat-globules. I cannot concur with Virchow in his opinion, that the strongly tinged epithelium of the tubes of Bellini contains fat, or that such cells are to be regarded as evidences of the existence of fatty degeneration.

*Origin.*—When we consider that vegetable food contains a greater or lesser quantity of fat, and that we find large quantities of the most ordinary vegetable fats accumulated in the animal organism, we might be disposed to infer that a vegetable diet was fully adequate to the nourishment of animals, since it has been discovered, or rather demonstrated, that it contains sufficient quantities of albuminous substances to compensate for the waste of the nitrogenous tissues. This view is daily confirmed by anatomical as well as purely physiological observations and experiments. Every farmer is well aware that cows will yield more butter when kept upon food abounding in fat, than when kept on fodder deficient in that ingredient, and that in rainy seasons, when plants contain less fatty matter, cows, although yielding large quantities of milk, give less butter than in dry seasons, although their food may be rich and good. If two organisms, similar in all respects, and under similar relations, partake of food, differing in its quantity of fat, there will be a difference in the deposition of fat. It cannot be doubted that a large portion of the fats passes from the food into the blood; we need only observe the chyle when the food has been of a fatty character, to convince ourselves, by the presence of fat-vesicles, that it has been converted into a perfect emulsion, whilst it will present only a slight turbidity from the presence of lymph- or colorless blood-corpuscles, when the food has contained but little fat. Boussingault<sup>3</sup> even succeeded, by

<sup>1</sup> Handwörterb. der Physiol. Bd. 2, S. 9.

<sup>2</sup> L'Experience, 1838. No. 42.

<sup>3</sup> Ann. de Chim. et de Phys. 3 Sér. T. 19, pp. 117–125, et T. 25, pp. 730–733.

a series of ingenious experiments, in showing that only certain quantities of fat passed in a given time from the intestinal canal into the general system, and that the excess of fat was discharged unchanged with the excrements. Thus he observed in the case of ducks, that a duck, when kept on the fattest food, could not assimilate more than 19·2 grammes of fat in twenty-four hours (or 0·8 of a gramme in one hour), from the *primæ viæ*.

A sharp contest has been obstinately maintained during the last ten years in reference to the question whether the animal organism does not possess the capacity of generating the requisite quantity of fat from other nutrient substances besides preformed fat. Dumas, Boussingault,<sup>1</sup> and some other French inquirers,<sup>2</sup> have endeavored to show by direct experiments, that herbivorous animals take up sufficient fat with their food, and that the animal organism has therefore no need of generating fat; while Liebig and his school<sup>3</sup> have arrived at a totally different conclusion from observations of a precisely similar character. For as they found that certain animals contained more fat, and discharged a larger quantity in their milk and excrements, than they had obtained by their food, they were led to the conclusion that the animal body must possess the property of forming fat from other organic substances. The contested point unfortunately long remained undecided, since the two parties differed in their idea of that which they termed fat in the food; the French inquirers regarding as fats all the matters that can be extracted from plants by ether, and Liebig reasonably enough considering those matters only as fats which possessed all other properties of fats besides that of solubility in ether. Liebig appealed in support of his views to the experiments first made by Huber, and afterwards repeated by Gundelach, and which appeared to prove that bees, when fed on pure sugar, are capable of generating wax. Subsequently, Dumas, in conjunction with Milne Edwards,<sup>4</sup> found reason to believe that bees cannot be fed for any length of time on pure cane-sugar; but that when fed upon the honey yielded by this sugar, which contains a very little wax, they were able to produce that substance. Boussingault,<sup>5</sup> Persoz,<sup>6</sup> and others, have since that period convinced themselves, by repeated experiments on pigs, ducks, and cows, of the correctness of Liebig's view, and therefore this long-contested question may now be regarded as at rest.

But it must not be forgotten that these experiments have only been conducted on the statistical method (that is to say, by a comparison of the quantity discharged and the quantity taken up by the organism); and that they cannot therefore afford more than the general demonstration that under many relations, fat must be formed within the animal body. But the following questions still remain unanswered: Does the animal body continue to exercise its property of generating fat, when a sufficient supply has been conveyed to it by food? What is the true seat of the formation of fat? And finally, how, and by what process,

<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. T. 12, p. 153.

<sup>2</sup> Persoz, in Compt. rend. T. 18, p. 245; Payen and Gasparin, in Compt. rend. T. 18, p. 797; Letellier, in Ann. de Chim. et de Phys. 3 Sér. T. 11, p. 433.

<sup>3</sup> Playfair, in Phil. Mag. Vol. 22, p. 281.

<sup>4</sup> Ann. de Chim. et de Phys. 3 Sér. T. 14, p. 400.

<sup>5</sup> Compt. rend. T. 20, p. 1726.

<sup>6</sup> Ibid. T. 21, p. 20.

and in what chemical proportion, is fat formed from starch or nitrogenous substances?

The first question, as to whether the organism *constantly exercises its power of forming fat*, does not admit of a solution in the present state of our knowledge, nor until a satisfactory answer can be given to the two other questions. If Boussingault's view be correct, that the ordinary vegetable substances contain sufficient fat to compensate for what has been lost through the functions of the animal body, we might infer that fat would only be generated from other substances when the food is deficient in fatty matters, or when the supply of fatty food is inadequate. It may, however, be argued against this teleological view, that if the conditions for the formation of fat are once present in the animal organism, this process will probably continue in operation without reference to the plus or minus supply of fat. But many pathological phenomena appear to show that this process may in some cases be abnormally excessive.

According to the views of Liebig and Scherer, in which most observers now concur, the *seat of the formation of fat* is to be sought in the *primæ viæ*. This hypothesis is not, however, based on strict proof, and its value greatly depends upon the origin we attribute to fat, namely, whether we derive it from albuminous, and therefore nitrogenous substances, or from starch, sugar, and other non-nitrogenous matters. Liebig's authority has given currency to the latter view, although it is opposed by many physiological facts. For if fat were formed in the *primæ viæ* from the starch of vegetables, the chyle would contain more fat after a vegetable than a fatty animal diet; but the contrary has invariably been noticed in all the observations made on this subject since the experiments of Tiedemann and Gmelin. Boussingault,<sup>1</sup> moreover did not observe any instance in his recent experiments on ducks, in which the fat contained in the intestinal contents, was increased by feeding the birds on starch or sugar, although such must have been the case if a metamorphosis of these substances into fat occurred in this part of the system. Thomson<sup>2</sup> was also led by his experiments on the influence of different kinds of food on the production of milk and sugar, to adopt the opinion that sugar had no part in the formation of fat. The occurrence of hydrogenous gases in the intestines, and the well-known fact of the reduction of alkaline sulphates into sulphides during the process of digestion in the intestinal canal, might indeed seem to afford some grounds for the possible reduction of the substances containing carbon and the elements of water, to which we apply the term carbo-hydrates, viz., starch, sugar, &c.; but until supported by some conclusive evidence, this view must be regarded as scarcely tenable in opposition to the facts referred to. H. Meckel<sup>3</sup> was indeed led to believe, from some experiments made on the subject, that sugar was thrown into a sort of fermentation by the bile, and was thus converted into fat; but it had escaped the attention of Meckel, who regarded every substance that dissolved in ether as a fat, that his ethereal extract contained not only fat, but all the products of decomposed bile soluble in ether; and the reason of his

<sup>1</sup> Compt. rend. T. 20, p. 1726.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 228-243.

De genesi adipis in animalibus. Dis. inaug. Hal. 1845.



obtaining a larger quantity of ether-extract when the bile was decomposed by sugar, than when digested without sugar, was simply in consequence of the presence of the sugar, which very much promotes the decomposition of the bile, and the formation of products easily soluble in ether (namely free biliary acids). It does not, therefore, appear from the facts already established, that fat is generated in the intestinal canal from sugar and starch, more especially as these substances would appear from Boussingault's experiments, to be too rapidly absorbed from the intestinal canal to allow of their being subjected to a fatty fermentation.

Liebig has advanced an hypothesis, that fat may also be formed from nitrogenous elements of food; and this view would appear to acquire support from the experiments made by Boussingault on ducks. For the latter observer found that when these birds had been fed on albumen and cascine, containing little or no fat, there was always more fat in their intestinal contents than when they had fasted for any length of time, or been fed only on clay, starch, or sugar. Unless, therefore, we would assume (which, indeed, we have no authority for doing), that fat is secreted in the intestinal canal after the use of nitrogenous substances, we must admit, from the above experiments, that a portion of fat may be generated in the *primæ viæ* from albumen containing no fat. It must, however, be observed, on the one hand, that the increase of the fat in the intestinal canal, after the use of albuminous food, is very inconsiderable, and on the other, that the experiments are so few in number, that we have not sufficient data for the satisfactory solution of so important a question. But it is very possible that the digestion of nitrogenous food may be accompanied by a greater secretion of bile than that of non-nitrogenous substances, and that the fats and products of decomposition of the bile, may have increased the ether-extract of the contents of the intestine, in the above experiments, after the use of nitrogenous food. As has been already observed, the solid excrements presented scarcely any *residua* of the bile except those which are soluble in ether.

Since the above facts do not, as yet, justify us in assuming that the seat of the formation of fat must be sought in the *primæ viæ*, we must turn to the processes at work in the blood, unless, indeed, we freely confess that nothing definite can, at present, be advanced on this subject.

The third question, as to *how* fat is formed from other substances, would next engage our attention, if the preceding considerations did not show that we are entirely deficient in the materials necessary for affording a satisfactory answer. For, so long as we are ignorant of the grounds on which a process is based, although we may be acquainted with its individual factors, we must defer all idea of a scientific explanation; there is, however, no deficiency of imaginary schemes to explain the formation of fat from sugar or protein. Support has been borrowed from the somewhat irrelevant fact of the butyric fermentation of sugar and starch; but as we have already observed (p. 42), there are no grounds for reckoning butyric acid among the fats, and the formation of metacetic, acetic, and formic acids, may just as well be regarded as processes of the formation of fat, as that of butyric acid. We are, therefore, for the present, constrained to regard this view as a mere fiction, illustrated

by chemical symbols, since, whatever corroboration it may acquire from future experiments, it is at present wholly devoid of all scientific support.

*Uses.*—We may regard the application of fat in the animal body as conducive to mechanico-anatomical, to physico-physiological, and chemico-physiological objects.

The uses of the fat deposited in the areolar tissue of the animal body are almost entirely of a strictly physical nature. If we reflect that fat is mostly found in a fluid state during life, we shall perceive some of the most useful properties which this condition imparts to the animal body. For, although fat is enclosed in separate layers and cells, it possesses so great a degree of mobility as to propagate pressure equally in all directions in the same manner as water. Every physicist knows that a bladder perfectly filled with water, cannot be brought to assume any given form without bursting; but we know that pressure applied to any part of such a body will be equally propagated in all directions. If, therefore, we suppose a number of such bladders to be laid side by side, enclosed in a larger space, and that we press one of them, the pressure thus applied will be propagated to all the others; and here we have an illustration of the uniform diffusion of external pressure through the whole adipose tissue. But besides the protection thus afforded the body from external shocks, it is further guarded in leaping and falling by the Haversian glands, which penetrate into the joints, and, receiving the shock, propagate it over a larger surface, by which its violence at each individual point must be very much diminished. Such was the object of nature in placing layers of fat on the soles of the feet, and the tuberosities of the ischium; and thus the depositions of fat were made to answer the purpose of water-cushions and other inventions of man's ingenuity, for the promotion of his ease and comfort.

Haller was the first who drew attention to the extreme utility of fat in filling up those interstices which must unavoidably exist between muscles, bones, vessels, and nerves. The bodies of children and women principally owe their rounded forms to the deposition of fat in the subcutaneous cellular tissue. The extreme mobility of the separate organs and parts of organs, is mainly owing to the same cause; and in every part of the body in which greater or less deposits of fat are met with, nature appears to have had a similar object in view. Hence fat is found to remain the longest in the parts where it is most needed, as in the heart, and in the orbit of the eye. How could so complicated a muscular structure as the heart move with freedom, ease, and regularity, if the interstices formed by the muscular bundles often contracting in opposite directions were not filled with fat, and if the vessels proceeding from them were not completely enclosed in fat? How would the muscles of the eye, and indeed the eye itself, act, if we could remove all the fat from the orbit of the living subject? Deprived of this protection, the muscles would become unable to discharge their functions, the optic nerve would be compressed, and sight utterly destroyed. Thus, too, we find in the rounded abdominal cavity, which is traversed by the cylindrical intestinal canal, that every fissure and interstice is filled up with fatty masses; in the great omentum, in the mesentery, and the *appen-*

*dices epiploicæ*,—wherever there is an interstice—we find fat; and it is most evident, that by these means all friction, and every violent shock, are diminished, while a free peristaltic movement is afforded to the intestinal canal. The lower part of the pelvis is especially furnished with fat of so yielding a nature as to permit of the organs of excretion contained in it, being dilated at will. How different would be the appearance of the face, if all the fat were removed from the muscles and from below the skin! The fat which smooths the bony corners and angles, and the narrow muscles of the face, is the cosmetic employed by nature to stamp the human countenance with the incomparable impress which exalts it far above all the lower animals. A similar physical use seems to be equally apparent in the deposition of fat on the extremities, although its presence may there be subservient to other purposes.

Although we find but little fat in the extremities of persons who are accustomed to exercise their muscles strongly, the quantity present is yet sufficient to effect the purposes already indicated.

Fat, when in a fluid state, is moreover a very *bad conductor of heat*. This property of fat has been most wonderfully employed by nature for the protection of the animal body from the injurious effects of excessive heat or cold, and of rapid alternations of temperature. Every one acquainted with the propagation of heat in fluid bodies, will easily perceive, that by the distribution of fat in small cells and layers, by which the rising and falling of the heated or cooled fluid is impeded, nature has most perfectly effected the object in view. We surround our stoves with stagnant air, in order to retain the heat as much as possible; but this object would be far more perfectly attained, if we could enclose the air in the subjacent and superimposed layers of so bad a conducting medium as the cellular tissue. When we consider the enormous quantity of cells filled with fat, which are frequently deposited under the skin of corpulent persons, we can scarcely comprehend how an otherwise healthy individual could die from the effects of excessive cold.

Thus we find that the whole abdomen is filled and covered with fat, for the purpose of maintaining that equable temperature which is requisite for the due performance of its various chemico-physiological processes, the adipose tissue of the omentum acting as a special protection to the abdominal viscera. In furtherance of a similar end, the female breasts are largely supplied with fat, since, from their exposed position, these organs might, without such a protection, readily become unfitted for their normal functions. The testicles, on the other hand, contain no fat, and the scrotum very little, because these organs must be kept cool, as we learn from the bad results following the non-descent of the testicles. Animal heat could not be maintained in so equable a condition in the body, if all the organs—every part in which a metamorphosis of tissue occurs—were not enveloped in fat. Do we not observe how eagerly phthisical patients, convalescents, and old persons, seek the warmth of the sun, and how emaciated animals delight in basking in its rays? We should probably also take into consideration the fact that, next to water, fat possesses the greatest capacity for heat, and hence a very considerable quantity of heat will be required to transmit warmth through the fatty investment of the body. As a proof that fat pos-

sesses these useful properties, we may refer to the practice common alike to the nations of the extreme north, and to the inhabitants of many tropical lands, of anointing the skin with fat, in order to guard in the one case against intense cold, and in the other against extreme heat.

The various uses arising from the low *specific gravity* of fat scarcely require comment. It would be almost impossible to swim without fat, and although it might be advanced that swimming is not a necessary faculty of the human body, we shall readily be disposed to admit the utility of fat in this respect when we consider that, if the muscles of only an arm were encompassed with pure water instead of fat, the force of the muscles, which is, moreover, better adapted to rapid movement than to overcome a resisting power, would undoubtedly be very considerably diminished; for there can be no doubt that in *hydrops anasarca* the muscular weakness does not depend alone on the tension, and on the morbid diminution of the muscular activity, but likewise on the altered condition of gravity of the whole extremity, depending on the accumulation of water and diminution of fat.

One of the best-known properties of fat, is that of its rendering other bodies *supple*, and diminishing as much as possible the brittleness of bodies, and the friction of parts moving on one another. This use is made most apparent in the movement of the muscles, and the free action of the joints. In this point of view, the utility of fat is nowhere more conspicuous than in the bones. Fat, undoubtedly, gives great flexibility to the earthy bones, as we perceive from their brittleness when macerated; and as is made most apparent in the disease of the bones inaptly termed osteomalacia, for, while there is so extraordinary a loss of osseous matter, that the bones appear, when macerated, to consist of a mere gauze-like tissue, most of the interstices are entirely filled with fat, as if the *vis naturæ medicatrix* would in some degree compensate, by an excessive accumulation of fat, for that property of the bones which has been destroyed by this disease.

I found, in the ribs of a patient who had died in a state of extreme osteomalacia, 56·92% of fat, together with 24·665% of other organic matters, 15·881% of phosphate, and 2·534% of carbonate of lime.

The utility of fat, considered in a mechanical point of view, is so evident from what has been already said, that it would seem superfluous to add any further remarks on the subject. If negative evidence were admissible, we might observe that fatty deposits are rarely or never found in the brain and lungs, where their presence would occasion mechanical injury, since external pressure, and even a slight increase of heat, would prove injurious to these organs. In the *glans penis* again we find no fat, because its presence would, undoubtedly, contribute to increase the irritability of this organ.

Before we proceed to the consideration of the chemico-physical uses of fat, we will cursorily advert to the view which has long prevailed in physiology, that the fat deposited in the areolar tissue is nothing more than a stored-up nutriment. This proposition, advanced in accordance with the earlier views of natural philosophy, appeared to derive a considerable degree of corroboration from a general consideration of the fatness and leanness of men and animals, under different physiological

or pathological relations; but such a method of observation is too vague and general any longer to maintain its ground in the present position of science. We have ceased to believe in the existence of a special administrator of the economy of the living organism, who, under the title of *vital force*, prepares, in times of plenty, for a season of scarcity; and we now know that the process of the deposition of fat in the areolar tissue is not so simple, and that its resorption does not admit of so ready a solution as was, at one time, believed to be the case. Thus, it must not be supposed that fat simply collects in the interstices of the cellular tissue, from which it may be as easily removed as the water which occasionally accumulates therein in *hydrops anasarca*. Fat is not contained in a free state within the interstices of the areolar tissue, but is contained in special cells, enclosed by an albuminous wall, and provided originally with a nucleus, the so-called cytoblast. Fat, therefore, only collects in the cellular tissue by means of a cell-formation, and hence it is, in many cases, extremely difficult to explain how fat can so rapidly disappear from the areolar tissue. It has not even been clearly determined whether the whole cell is resorbed with the fat, or whether, as Gurlt<sup>1</sup> maintains, the cell remains, and is filled with serum instead of fat. We must remember, in considering the observations made on the increase or diminution of fat in men and animals, in a healthy as well as a diseased condition, that fat-cells, like most other animal cells, stand in a constantly alternating relation to the other fluids, more especially the blood. The constitution of the blood is reflected in all parts of the animal body, and endosmotic and counter currents must be established as soon as one of the fluids in question is subjected to any alteration. It is not necessary that we should assume with Mascagni, that each fat-cell is provided with an artery and a vein, for the relations of endosmosis with which we are at present acquainted sufficiently explain the different results of this mutual action between the nutrient fluid and the fat-cell. In rapid emaciation, and more particularly in those conditions of the body which are usually termed anæmic (as, for instance, after repeated bloodletting and other losses of the animal fluids, and after typhus and other severe diseases), fat is often accumulated in the blood, while it disappears from the subcutaneous cellular tissue. Conversely, the formation of fat-cells often appears to be more rapid than the reproduction of other tissues after anæmic conditions, when the blood has not quite recovered its normal character; hence we frequently observe a very abundant deposition of fat after typhus and other diseases resulting in anæmia. We shall enter more fully into the consideration of this subject, when we proceed, at the close of the physiological chemistry, to treat of the general phenomena of nutrition.

We now enter upon what may be termed the physico-physiological uses of fats. Liebig has shown, with his characteristic ingenuity, that the fats mainly contribute to *the excitement and maintenance of animal heat*. One of the most ingenious of Liebig's deductions is his classification of the elements of nutrition into true plastic nutrient substances and food for the respiration, to the latter of which he especially ascribes the functions of maintaining animal heat. But as, in our observations on the

<sup>1</sup> Physiol. S. 20.

processes of respiration and nutrition (in the second volume), we shall enter more fully into the examination of Liebig's views on this subject, we shall here only observe that, however paradoxical and apodictic many of his deductions may appear, he has founded a new era in physiological chemistry, and has been the means of throwing a clearer light over the whole economy of the organism. Owing to his aphoristic mode of representation, his views have often been misunderstood and erroneously interpreted, and many persons have even supposed that they must assume that fat is simply transferred into the blood, where it is burned like the oil in a lamp, or the coke in a steam-engine. A more attentive examination of Liebig's writings shows, however, that he did not entertain so crude a view of the subject. But we must admit that we do not consider as wholly groundless the objection which has been advanced against Liebig, that he regards animal heat as too independent of other processes. Animal heat can only be considered under one of two points of view; that of being an incidental phenomenon and the mere result of certain vital processes, or as being necessary to the maintenance of definite animal processes and functions. If the latter view be even partially correct, we must recollect that animal life is not generally dependent upon a definite high temperature, and that numerous cold-blooded vertebrate animals perform the processes of digestion, respiration, blood-formation, and of the nervous system, as well at a low temperature, as warm-blooded animals do at  $37^{\circ}\cdot5$ . If, on the other hand, animal heat were a mere incidental phenomenon, the fats would appear to be most uselessly expended in serving no other purpose than that of developing heat. The fat of the living body therefore probably conduces to other ends in the animal economy.

I was long since led, from theoretical grounds, to regard the fat as one of the *most active agents in the metamorphosis of animal matter*; and this subjective conviction has since been converted into objective proof by numerous experiments and observations. After having found by experiments regarding the fermentation of milk,<sup>1</sup> that this process cannot be excited by albuminous bodies in saccharine or amylaceous fluids, excepting with the co-operation of fat, I next ascertained that a certain, although small quantity of fat, was indispensable to the metamorphosis and solution of nitrogenous articles of food during the process of gastric digestion. Elsässer<sup>2</sup> has confirmed the fact by the observation that, in experiments on artificial digestion, the solution of articles used as food is considerably accelerated by means of fat. It is easy to ascertain by means of artificial openings in the stomachs of dogs, that flesh containing only little fat, and especially albuminous substances which have been designedly deprived of their fat, remain longer in the stomach, and therefore require a longer period for their metamorphosis, than the same substances when mixed or impregnated with a little fat. An excess of fat appears, on the other hand, at least in persons of weak digestion, to exert an injurious action. The pancreatic juice most probably owes a portion of its utility in promoting digestion to the quantity of fat which it contains.

The pancreatic juice, like pus, deposits, according to Cl. Bernard,<sup>3</sup>

Simon's Beiträge. Bd. 1, S. 63-77.

<sup>2</sup> Magenerweichung der Kinder. S. 112.

<sup>3</sup> Arch. gén. de Méd. 4 Sér. T. 19, p. 71.

fine crystalline bundles of margarin and margaric acid during its spontaneous decomposition at a high temperature.

Although we are unable fully to demonstrate the special agency of fat in the further metamorphosis of the digested food, namely, in the formation of chyle and blood, yet we need only observe the intestinal villi during the process of digestion, and see their individual cells filled either with clear fat or dilated by a grumous matter—we need only institute a microscopic and chemical comparison of the fat in the chyle found in the finest lacteals with the contents of the thoracic duct, in relation to the different quantity and character of the fat in both fluids—in order to perceive that fat is not only resorbed, but that it also influences the metamorphosis of the albuminous constituents of the nutrient fluid. Is it probable that fat would so tenaciously adhere, even under different modifications, to some of the constituents of the blood, unless it exercised some influence on their origin or metamorphosis? Or are we to suppose that the fat, which we can extract from the animal nerves by boiling them with alcohol, or digesting them with ether, and whose removal leaves the separate nerve-fibres like hollow cylinders with thick walls, is deposited there for no useful end, and that it can be wholly free from all co-operation in the function of the nervous system?

However opposed we may be to teleological explanations, we cannot deny the importance of an inquiry into the grounds and aims of obscure subjects, since it is by such means that natural inquiry has ever been guided into those paths which lead to the investigation of causes, and the final comprehension of phenomena.

We have already become acquainted with two species of animal cells, in which fat is the main constituent, viz., true fat-cells and certain kinds of granular cells (the so-called inflammatory globules) found in milk (*Corps granuleux*, *Colostrum-corporuscles*), in the sputa in chronic catarrh, in old apoplectic cysts, &c. Fat, however, would appear from some of the latest investigations of the most distinguished physiologists, to play a very important part in every kind of cell-development; indeed most inquirers agree in regarding it as affording the primary foundation in the formation of a cell. Acherson<sup>1</sup> was undoubtedly the first to direct attention to this subject by his discovery that albumen always coagulates around a fat-globule placed in an albuminous solution; and although the question may not be so simple as Acherson would make it appear, the presence of fat in the cell during its formation, and its importance in affording the predisposing cause of cellular formation, is no longer denied by any physiologist, whether he adhere to the old theory of cell-development established by Schwann and maintained by Kölliker, or advocate the views of Henle, or of Reichert. According to Hünefeld, Nasse, and others, the nucleoli invariably consist of fat. The newly secreted or recently formed plasma always contains more free fat than after the nuclei or cells have been deposited,—a fact that is clearly de-

<sup>1</sup> Müller's Arch. 1840. S. 49. [In connection with this subject, I may refer to a Memoir on "The Structural Relation of Oil and Albumen in the Animal Economy," read by Professor Bennett before the Royal Society of Edinburgh, and published in the "Monthly Journal of Medical Science," Vol. 8, p. 166; a Lecture published by myself in the "London Medical Gazette," for May, 1848, New Series, vol. 6, p. 140; and v. Wittich, Ueber die Hymenogonie des Eiweisses. Königsberg, 1850.—a. e. n.]

monstrated in H. Müller's<sup>1</sup> excellent memoir on the chyle and its histological elements, who shows that the cloudy turbidity of the chyle which depends on the presence of the fat, disappears in proportion as the isolated granules, the aggregated granules, and the cells are developed. The serum of pus moreover contains much less fat than pus-corpuscles. In the blood we find that fat is especially deposited in the cells and in the fibrin, the granular contents of many of the blood-corpuscles consisting of this substance. All plastic exudations contain more fat than the non-plastic; for the latter, as dropsical fluids and tubercular masses, although occasionally containing much cholesterin, usually contain very little true fat; while on the other hand exuberant, highly cellular cancers abound in this ingredient.

In pus, the pus-corpuscles often sink some lines below the level of the fluid; on comparing the amount of fat in the supernatant serum with that in the pus beneath it in which the corpuscles were suspended, I observed, in two experiments conducted with different pus, that in one there was only 7.13% of fat in the solid residue of the serum (which should have contained most of the fat since it was taken from the surface of the pus after it had stood a long time), while the thick purulent sediment contained 18.41%; in the other case there was 9.084% in the residue of the serum, and 17.14% in that of the pus. The difference between the amount of fat in the serum of the pus and in the pus-corpuscles is most plainly apparent when both the sediment and the serum of good pus are suffered to remain in well-closed vessels. Both fluids become acid, and fats and fatty acids are separated from them; in the former these changes are but slightly developed, whilst the acid purulent sediment exhibits, under the microscope, an innumerable quantity of the most beautiful crystallizations of margaric acid and of margarin, with cholesterin.

The fats of the blood are also principally deposited in the cells or blood-corpuscles. I found in 100 parts of well-dried blood-corpuscles taken from the blood of the ox, and whose mode of preparation I shall explain in another portion of this work, 2.214% of fat in one experiment, and 2.284% in another; the fibrin of the same blood contained in the one instance 3.218%, in the other 3.189% of fat; while 100 parts of the solid residue of the serum yielded 1.821, and 1.791 parts of fat. The blood-corpuscles have, unfortunately, scarcely ever been examined with reference to their amount of fat; in other respects, however, a comparison with the analyses instituted by other observers on the blood, leads to the same result.

It may be observed, in reference to the small quantity of fat contained in tubercles, that many fat-vesicles are often discovered under the microscope in recent tubercular deposits, as, for instance, in gelatinous tubercles, but that gray, solid tubercles, when submitted to a chemical analysis, after the separation of the cholesterin, which although not belonging to the fats is always reckoned amongst them, are found to contain very little fat. In a gray tubercular mass, I once discovered only 3.54% in the well-dried substance, although almost every other

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 2, S. 238.



tissue contained far more fat. Becquerel and Rodier<sup>1</sup> found, moreover, that in tuberculosis the saponified fats were far more diminished in the blood than in any other fluid.

We may here, perhaps, find some explanation of the mode of action of cod-liver oil, whose utility cannot be wholly denied even by that spirit of scepticism which has of late been so prevalent in medicine; and we have always been of opinion that cod-liver oil acts upon certain stages of disease more by its true fatty nature than by the small quantity of iodine which it contains. In confirmation of this view we may observe that many experienced practitioners (Oppolzer among the number) have found that almond oil and other similar oils are as efficacious as the loathsome cod-liver oil. But the idea that cod-liver oil, considered (according to the misconception of Liebig's views<sup>2</sup>) as a mere material of combustion, should be of benefit in a disease where the lungs are so entirely clogged or degenerated that an extensive oxidation of the blood is impossible, can only be entertained by persons wholly ignorant of the character of tuberculosis or of pulmonary consumption. No chemical analysis is needed to show that cellular cancer (encephaloid) and sarcoma abound in fat, and every one who has examined one or two of such tumors microscopically will be able to confirm the truth of this ordinary observation.

When we consider all these facts we shall be almost involuntarily led to the conclusion that fat takes a highly important share in the most important, and at the same time the most mysterious processes in the formation of cells and tissues. We cannot believe that fat is a mere incidental agent in all these processes, but we must rather regard it as of essential aid in the process of converting nitrogenous nutrient substances into cells and masses of fibres, in like manner as it co-operates in the processes of lactic fermentation and digestion; and it is probable that whenever a chemical equation representing the formation and function of certain cells can be established, fat will constitute one of the integral factors. Indeed it is impossible to believe that in the vital activity of cellular action, fat should be without influence on the metamorphosis of the substances which it accompanies, and that without reference to them, it should obey only its own affinities towards oxygen or an alkali.

In considering fat as an important agent in the various phases of the metamorphosis of animal matter, we cannot, however, refer its action solely to mere contact or a catalytic force, but we are constrained to assume that it co-operates in the metamorphic action, and experiences metamorphoses, combinations, and decompositions. None but those chemists, who, imagining they comprehend Liebig's views, have framed and illustrated a physiology of their own, in the same manner as speculative natural philosophers have attempted *à priori* to construct the laws of the natural sciences, could have regarded the animal body as a furnace, and fat as a simple and crude material of combustion. It is, however, the province of physiological chemistry to trace the chemical phenomena of the animal body and its various substances in their separate phases of metamorphosis, and from the knowledge thus obtained, to sketch the grand and universal features of chemical action in the

<sup>1</sup> Gaz. Méd. 1844. No. 51.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 84-89.

living body. It would be equally unphysiological and unscientific to suppose that the requirements of physiology would be fully satisfied by our proving that fat becomes finally decomposed into carbonic acid and water. The province of physiological chemistry is rather to show whether fat, or rather the fatty acids, always gradually and successively lose two atoms of carbo-hydrogen, that is to say, whether remaining in accordance with the general formula, they become converted into acids of the first group, and are then finally decomposed into carbonic acid and water; or whether fats contribute by their metamorphosis in the animal body to form other known animal substances. As, however, in the present state of our positive knowledge, we are unfortunately not in a position to answer this question with certainty, it is better to confess our ignorance, than to indulge in vague conjecture, although many chemical and physiological experiments afford some support to the hypothesis, that the fats take a part in the formation of other substances which cannot be regarded as mere products of their oxidation.

Since we find so large a quantity of saponified fats in the blood and other animal fluids, as for instance in the bile, it is not improbable that the first step in the alteration of the fats consists in their decomposition into glycerine and the corresponding fatty acids. If we assume that the fats are subjected to so gradual an oxidation that their carbo-hydrogen radical gradually diminishes by 2 atoms of carbo-hydrogen, it is singular that we should find the fatty acids which mark the gradations from capric to margaric acid in plants, but not in animals; for while the formation of fatty acids with a high atomic weight is very gradual in plants, a similar law does not prevail in reference to their regressive formation in animals, for here we meet with no acids besides margaric and stearic having a fat-radical of the formula,  $C_nH_{n-1}$ . It would appear, therefore, that the fatty acids, when separated from glycerine (to which reference has already been made at p. 218) enter into complicated combinations and metamorphoses, in which it is not easy to recognize or detect their presence. We have already (at p. 120) noticed the probability that the principal acid contained in the bile, cholic acid, is a conjugated fatty acid; chemical experiments giving evidence of the presence of oleic acid in it, although it cannot actually be separated.

The hypothesis, that *a portion of the fat takes part in the formation of bile*, is further confirmed by numerous physiological and pathological experiments.

The following physiological facts in some degree confirm this view. A close observation of the development of the chick within the egg, leads us almost irresistibly to the opinion, that towards the close of the period of incubation, a portion of the fat in the yolk-sac (when it is drawn into the abdominal cavity and adheres to the liver) is converted into biliary matter; and every physiological inquirer, who has occupied himself with this subject, must have observed the greenish tint which is often, although not always, very distinctly visible in the yolk-sac, and especially along the course of the veins. On one occasion I found this color so intense, that I was induced to treat the whole of the yolk-sac and its contents with boiling alcohol, and examine it for bile, according to the method described at p. 118; when the ordinary bile-reaction was

obtained by Pettenkofer's test. The veins of the yolk-sac pass into the liver, and it is well known that the vessels of the yolk-sac for the most part resorb the yolk, and transfer it into the liver; for the earlier view that the yolk passes through the *ductus vitello-intestinalis* into the intestine, and is carried from thence into the liver by the biliary ducts, is incorrect. The liver at this period serves mainly, as E. H. Weber,<sup>1</sup> and Kölliker<sup>2</sup> have shown, to form colorless and colored blood-corpuscles, and not to produce or secrete bile, for I have frequently convinced myself by observations on human and animal embryos, that at this period the gall-bladder contains no bile.

The *blood of the portal vein*, from which the bile is principally formed, differs from all other blood, whether venous or arterial, by its large quantity of fat, as was noticed by Simon and Schultz, and has been corroborated more recently by the exact quantitative analyses of Fr. Chr. Schmid,<sup>3</sup> who found that the blood of the portal vein contained so much more fat than that of the jugular vein, that he was led to regard this as the most essential difference between these two kinds of blood. Moreover he observed that the fat from the blood of the portal vein was of a dark-brown color, and that it was always richer in olein, and consequently more greasy, than the fat of other venous blood, which is white and crystalline. When animals are *starved* for any length of time it is well known that they rapidly become emaciated; the urine still exhibits nitrogenous constituents, corresponding in amount to the products of effete tissue; whilst the gall-bladder is perfectly full, and the liver constantly pours forth bile into the intestine, as I have convinced myself by a repetition of Magendie's experiments.<sup>4</sup> The above fact seems to explain the cause of the bitter taste of which persons suffering from starvation very frequently complain. Whence can the liver extract the materials necessary to the formation of bile? The urine, although poorer in solid constituents, always contains a considerable quantity of urea; and the animal body contains few or no highly carbonaceous substances, with the exception of fat, which we here observe disappearing very rapidly, while at the same time there is an abundant secretion of bile. The experiments of Bidder have, however, most distinctly proved, by the most careful determination of the excreta in fasting animals, that the elements excreted by the lungs and kidneys cannot solely originate from nitrogenous tissues, but that the excess of carbon and hydrogen excreted by the lungs is entirely to be referred to the decomposition of the fat of the starving animal, especially since these determinations of the excreta exactly coincide with the loss of fat directly observed in the dead body of the animal. The daily diminution of the biliary secretion in fasting animals occurs in almost the same ratio as the loss of fat in the body (Schmidt).<sup>5</sup>

In *disease*, the diminution or increase of fat is inversely proportional to the secretion of bile. Polycholia, which seldom occurs in adults, but which in children constitutes the affection known as *Icterus neonatorum*,

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 4, S. 160-164.

<sup>2</sup> Ibid. Bd. 4, S. 112-160.

<sup>3</sup> Heller's Arch. Bd. 3, S. 487-521, and Bd. 4, S. 15-37, and S. 97-132.

<sup>4</sup> Journ. de Physiol. T. 8, p. 171.

<sup>5</sup> Verdauungsäfte u. Stoffwechsel. Mitau, 1852, S. 386-398.\*

is always accompanied with rapid emaciation. In acute diseases, emaciation generally occurs in conjunction with critical symptoms, that is to say, when the organs of excretion resume their activity, and eliminate the materials that have become effete; hence the copious semi-solid fæces. In all acute or chronic diseases of the liver, the fat accumulates either merely in the blood, or in the blood and in the cellular tissue. The obesity observed in habitual drunkards is not in consequence of their taking too much combustible material into their bodies (brandy drinkers moreover generally take only small quantities of solid food), but in consequence of the disturbed hepatic action, which the invariably abnormal condition of the liver, found after death in these cases, proves to have existed.

Traill<sup>1</sup> and Lecanu have found the blood extremely rich in fat in inflammation of the liver; and Lassaigne,<sup>2</sup> and more recently Becquerel and Rodier, found the quantity of the fat in the blood more increased in icterus than in any other disease. Dobson, Rollo, and Marcet, observed so large a quantity of fat in the blood of diabetic patients that it resembled an emulsion; but I have myself only on two occasions found the blood to be largely charged with fat in diabetes, and here the disease was complicated with an affection of the liver, and the excrements of the patients were pale, and almost of a grayish-white tint.

All these facts render it difficult to deny the existence of a connection between fat and the formation of bile.

It is not, however, wholly impossible that fat should contribute in some measure to the formation of other substances, but we will here simply observe that facts subsequently to be noticed give some probability to the opinion that fat likewise co-operates in the *formation of the blood-pigment*.

We trust that the above remarks will lead to a more careful inquiry into the metamorphoses and function of fat in the healthy and diseased body, and be the means of assigning a higher degree of importance to this substance, than has hitherto been awarded to it in the animal economy.

#### HYDRATED OXIDE OF CETYL.— $C_{32}H_{33}O.HO$ .

##### *Chemical Relations.*

*Properties.*—This substance, to which its discoverer, Dumas, gave the name of *ethal*, forms white, solid, crystalline plates, melts at about  $56^{\circ}$ , again solidifies at  $48^{\circ}$ , and volatilizes readily either alone or with aqueous vapor, when heated; it is devoid of smell and taste, is insoluble in water, but dissolves in all proportions in hot alcohol and ether, has no action on vegetable colors, and when ignited burns like wax. It is decomposed when heated with nitric acid; heated to  $220^{\circ}$  with hydrated potash, it becomes converted (see p. 75) into cetylic acid ( $C_{32}H_{33}O + 2HO + KO = 4H + KO.C_{32}H_{31}O_3$ ). When warmed with concentrated sulphuric acid it forms an acid haloid salt.

<sup>1</sup> Annals of Philos. 1829, vol. 5, p. 199.

<sup>2</sup> Journ. de Chim. Méd. T. 2, p. 264.

*Composition.*—According to the above formula, deduced from the analyses of Dumas and Peligot,<sup>1</sup> this body consists of:

Carbon,	.	.	.	.	.	32 atoms,	.	.	.	79.889 *
Hydrogen,	.	.	.	.	.	33 "	.	.	.	18.636
Oxygen,	.	.	.	.	.	1 "	.	.	.	3.306
Water,	.	.	.	.	.	1 "	.	.	.	3.719
										<hr/> 100.000

The atomic weight of the hypothetical anhydrous substance = 2912.5.

This body, like glycerine, is the hydrate of a fatty base; but its composition and its relations of combination indicate that it is much more closely allied to the hydrated ethers or alcohols; in common with them it is included in the formula  $C_nH_{n+1}O.HO$ , it loses the one atom of water in combining with acids, and is converted by oxidation into an acid of the formula  $C_nH_{n-1}O_3$ . Oxide of cetyl or cetylic ether in an anhydrous state has not been obtained.

*Combinations.*—Very little is known of the *acid sulphate of oxide of cetyl* in its isolated state. Its combination with potash, which =  $C_{32}H_{33}O.SO_3 + KO.SO_3$ , is obtained in minute, thin, nacreous plates.

*Cetylate of oxide of cetyl*,  $C_{32}H_{33}O.C_{32}H_{31}O_3$  (Smith<sup>2</sup>) exists preformed, under the name of *cetin* or *spermaceti*, principally in the cavities of the skull, but also in the fat of other parts, of the *Physeter macrocephalus*. To obtain it in a state of purity, we must repeatedly crystallize it from hot spirit, of 0.816 specific gravity. It separates in minute, nacreous, white plates, devoid of smell and taste; it fuses at 49°, but on cooling solidifies in a crystalline form; it volatilizes at 360° without decomposition, dissolves in 40 parts of boiling spirit, of 0.821 specific gravity, and more readily in anhydrous alcohol and ether; when submitted to dry distillation it yields no pyroleic acid, and when digested with nitric acid it yields adipic but no suberic acid. When heated with hydrated potash it is resolved into hydrated oxide of cetyl and cetylic acid.

*Preparation.*—In order to obtain hydrated oxide of cetyl, pulverized hydrated potash must be added to melted spermaceti, and the mixture be continuously stirred; when the mass has become solid it must be digested with water, and the soap which is thus produced must be treated with hot dilute hydrochloric acid; after the oily stratum has been again fused with caustic potash, and digested with hydrochloric acid in order to insure the perfect decomposition of the cetin, the mixture of cetylic acid and oxide of cetyl must be digested with milk of lime and evaporated. From this mixture we can take up the hydrated oxide of cetyl by cold alcohol; which does not dissolve the cetylate of lime.

*Tests.*—It is impossible to recognize this substance with certainty unless by an elementary analysis.

#### *Physiological Relations.*

Hydrated oxide of cetyl has not yet been found in an isolated form; spermaceti, however, occurs in several parts of the Cachalot, mixed with ordinary fat; in greatest quantity, however, in the head, not in the

<sup>1</sup> Ann. d. Chim. et de Phys. T. 72, p. 5.    <sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 42, S. 40-51.

actual cavity of the cranium, but in a large excavation on either side of the upper part of the head and lying external to the nostrils. Regarding the formation and uses of this substance, we can only offer the same opinions as respecting the fats in general.

The *doeglic oxide* of Scharling is too hypothetical a body to be entitled to be yet classed among the haloid bases. Compare p. 111.

## LIPOIDS.

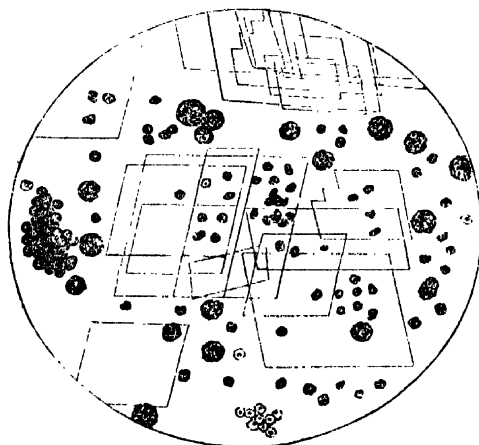
Under this head we place what are termed the non-saponifiable fats, that is to say, such bodies as have many physical properties in common with the salts of oxide of lipyl, but do not resemble them in their composition or in their products of decomposition, and consequently cannot be placed amongst the true fats. In this class we place *cholesterin*, *serolin*, *castorin*, and *ambrein*.



### *Chemical Relations.*

*Properties.*—This body, formerly known as *biliary fat*, separates from its solutions in nacreous scales containing 2 atoms of water; examined under the microscope, these crystals appear in very thin rhombic tablets,

Fig. 16.



Cholesterin.

whose obtuse angles =  $100^{\circ} 30'$ , and whose acute angles =  $79^{\circ} 30'$ ; it fuses at  $145^{\circ}$ , solidifying again, and becoming perfectly crystalline at  $135^{\circ}$ ; it may be distilled *in vacuo* at  $360^{\circ}$  without undergoing decomposition; it becomes electrical on friction, is perfectly insoluble in water, but dissolves in 9 parts of boiling alcohol, from which the greater part

again separates on cooling; it is also slightly soluble in soap-water, and more freely in the fatty oils and taurocholic acid; it is inflammable, and burns with a smoky flame. Treated with concentrated sulphuric acid it assumes a red tint at  $60^{\circ}$ , and is converted, with the loss of water, into three probably polymeric carbo-hydrogens, which their discoverer, Zwenger,<sup>1</sup> has named *cholesterilins*. If cholesterin be heated with concentrated phosphoric acid to its melting-point, there are formed two carbo-hydrogens, isomeric with the cholesterilins, to which Zwenger<sup>2</sup> has applied the name of *cholesterones*. By prolonged boiling with concentrated nitric acid, it becomes first converted into a resinous mass, which, by prolonged digestion, is resolved (Redtenbacher<sup>3</sup>) into acetic, butyric, caproic, oxalic, and cholesteric acids (see page 117). A portion of the hydrogen may be abstracted from cholesterin by chlorine or bromine, of which an equivalent quantity takes the place of the hydrogen thus removed. It is not decomposed by concentrated alkalis, even when the mixture is submitted to prolonged heat. On dry distillation it leaves a charcoal, and yields an oily distillate, which after rectification with water evolves an agreeable odor, resembling that of the Geranium.

*Composition*.—Cholesterin has been analyzed by Marchand,<sup>4</sup> Schwendler and Meissner, and subsequently by Payen,<sup>5</sup> with tolerably similar results, which have led to the establishment of the above formula,  $C_{37}H_{72}O$ . As we have not yet succeeded in combining cholesterin with any other body, we have no means of controlling this formula and of determining its atomic weight. Zwenger has very recently analyzed the *cholesterilins*, of which he was the discoverer, and found them composed in a tolerably uniform manner. He assumes, however, that there are differences between them, and that they may be respectively represented by  $C_{32}H_{26}$ ,  $C_{22}H_{18}$ , and  $C_{27}H_{22}$ ; and he believes that cholesterin consists of these three carbo-hydrogens and 3 atoms of water, its formula being, according to his views,  $C_{81}H_{69}O_3$ . Taking into consideration the limited accuracy which we are capable of attaining in our elementary analyses, and the method by which we deduce a formula from empirical results, we must regard Zwenger's view as, at present, very hypothetical.

We give the composition of cholesterin according to both formulæ :

Carbon,	.	.	.	.	37 atoms,	84.733	81 atoms,	83.93
Hydrogen,	.	.	.	.	82 "	12.214	69 "	11.91
Oxygen,	.	.	.	.	1 "	3.053	3 "	4.16
						100.000		100.00

Notwithstanding its extraordinarily high numbers, Zwenger's formula accords more closely than the simpler one with the empirical results.

*Products of decomposition*.—*a. Cholesterilin*,  $C_{32}H_{26}$ , is earthy, amorphous, insoluble in water, and slightly soluble in alcohol; it differs from the two other carbo-hydrogens by its insolubility in ether; it crystallizes from a hot oil of turpentine solution, and melts and is decomposed at  $240^{\circ}$ .

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 66, S. 5-13.

<sup>2</sup> Ibid. Bd. 69, S. 347-354.

<sup>3</sup> Ibid. Bd. 57, S. 162-170.

<sup>4</sup> Journ. f. pr. Ch. Bd. 16, S. 37-48.

<sup>5</sup> Ann. de Chim. et de Phys. 3 Sér. T. 1, p. 54.

*b. Cholesterilin*,  $C_{22}H_{38}$ , crystallizes in minute, strongly glistening plates or delicate needles, which are insoluble in water and alcohol, but soluble in ether; it fuses at  $255^{\circ}$ , and on cooling solidifies in a crystalline form.

*c. Cholesterilin*,  $C_{27}H_{44}$ , is a yellow, amorphous, resinous mass, freely soluble in ether, slightly so in alcohol, and insoluble in water; it fuses at  $127^{\circ}$ . Both this and the preceding variety are decomposed by heat. The formulæ must be regarded as entirely hypothetical, since the percentage composition, both as found and as calculated, approximates to 88% of carbon, and 12% of hydrogen for all three of them.

*a. Cholesterone* is obtained by extracting with spirit the residue of cholesterin, heated with phosphoric acid to  $137^{\circ}$ ; it crystallizes in right rhombic, bilaterally acuminate prisms; is colorless, transparent, very lustrous, lighter than water, and melts at  $68^{\circ}$  into a colorless fluid, which very slowly reassumes the solid form; it can be distilled for the most part undecomposed, and burns with a smoky flame. It is insoluble in water, but dissolves freely in alcohol and ether, and in the volatile and fatty oils.

*b. Cholesterone* is extracted by ether from the residue insoluble in alcohol; it occurs in fine white needles, melts at  $175^{\circ}$ , cannot be distilled without partial decomposition, is lighter than water, is devoid of taste and smell, and burns with a smoky flame. Both varieties of cholesterone are devoid of oxygen, but contain about 12 parts hydrogen to 88 of carbon.

*Preparation.*—The best method of preparing cholesterin is by boiling gall-stones, containing this substance, with alcohol, and filtering the solution while hot; by recrystallization from hot alcohol it is easily obtained in a state of purity.

*Tests.*—The recognition of cholesterin in the animal fluids is by no means so easy as might be supposed from the distinctive characters of this body; if, however, it has been once separated in a crystalline form, nothing is easier than to diagnose its presence with certainty. If, by its insolubility in water, acids, and alkalies, and by its solubility in alcohol and ether, it has been recognized as a fatty substance, it may be readily distinguished from all other similar substances by a measurement of the angles of the rhomb. It is only necessary to remark that the tablets are often so thin that their contour may be easily overlooked in a microscopic examination, if other morphological substances are simultaneously present in the field of the microscope: we must then slightly shade the field by a lateral or central diaphragm to make the outline stand forth more distinctly. In all this there is no difficulty; but it is, on the other hand, often very troublesome to obtain this substance in a crystalline form from oily fluids containing bile, or from soapy solutions. If we saponify with an alkali the fat which holds the cholesterin in solution, it also dissolves in the soap-water, and on the addition of an acid is again converted into the fatty acid; hence, when dealing with very small quantities of cholesterin, it is necessary to combine the fatty acid with oxide of lead, and to extract with boiling alcohol; the small quantity of dissolved margarate of lead is usually deposited previously to the separation of the cholesterin, which frequently does not crystallize, so as to



be recognized by the microscope, until the fluid has been submitted to evaporation.

### *Physiological Relations.*

*Occurrence.*—Small quantities occur in most of the animal fluids. It was originally discovered by Gren in *biliary calculi*, and has since been recognized as a constant ingredient of the *bile*. In the normal condition cholesterol is dissolved in the bile, and hence cannot be recognized under the microscope; even in the bile removed from the dead body we rarely find tablets of cholesterol (Gorup-Besanez),<sup>1</sup> and in these cases we cannot tell whether it depends on an augmentation of the cholesterol or on its separation in consequence of the decomposition of taurocholic acid. Frerichs<sup>2</sup> found no cholesterol in several examinations which he made of the bile in cases of fatty liver.

Cholesterol was first distinctly recognized as a normal constituent of the *blood* by Lecanu, Denis, Boudet, and Marchand; while Becquerel and Rodier<sup>3</sup> have especially directed attention to its augmentation and diminution in diseased conditions of the blood. According to these authors the amount of cholesterol in 1000 parts of normal blood ranges from 0.025 to 0.200 (the mean being 0.088). There is an augmentation of the cholesterol in the blood in old age, and in most acute diseases soon after the occurrence of febrile symptoms, especially in inflammations and in icterus. They have not discovered any physiological or pathological condition in which there is a constant diminution of this substance.

Cholesterol always occurs in the *brain*, where it was first discovered by Couërbe. Many subsequent observers have confirmed his observations.

It also appears to be an integral constituent of *pus*; at least whenever I have allowed pus to become sour I have found tablets of cholesterol in the decomposed mass; moreover, Caventou, Güterbock, Valentin, and many others, have detected it in fresh pus.

Cholesterol is also very frequently found in dropsical exudations, especially in cysts; I have recently, on two occasions, analyzed the *fluid of hydrocele* discharged by incision; both specimens were semi-solid rather than fluid, and when stirred, formed beautiful glistening bands. Their only morphological element was cholesterol.

*Obsolete (chalky) tubercle*, old *echinococcus-cyst*, such as are often found in the liver, and degenerated *ovaries* and *testicles*, often contain a large amount of cholesterol.

I once found the *choroid Plexus* in the brain perfectly incrustated with cholesterol.

In *encysted tumors* (especially in meliceris) as well as in *carcinomatous* and *other tumors*, we often meet with cholesterol.

In the *solid excrements* we may generally recognize traces of cholesterol; and in the *meconium* this substance is present in very considerable quantity.

In *pulmonary expectoration* I have only found cholesterol in the cheesy

<sup>1</sup> Untersuchungen üb. Galle. Erlangen, 1846. S. 58.

<sup>2</sup> Hannov. Ann. Bd. 5, H. I.

<sup>3</sup> Gaz. Méd. 1844, No. 47.

concretions ejected in advanced phthisis, and when vomicae are already present.

In the *urine*, as far as I know, no cholesterin has yet been found.

[Möller states that he has twice discovered cholesterin in the pellicle which forms on the urine during pregnancy, but I know nothing of his character as an observer. See Casper's *Wochenschr.* 1845, N. 2, 3; or my translation of Simon's *Animal Chemistry*, vol. 2, p. 333.—G. E. D.]

*Origin.*—In regard to the origin of cholesterin, which is never found in the vegetable kingdom but only in the animal body, we cannot offer even a probable conjecture. Judging from the mode of its occurrence, we must regard it as a product of decomposition; but from what substances and by what processes it is formed, it is impossible even to guess. Notwithstanding the similarity which many of its physical properties present to those of the fats, we can hardly suppose that it takes its origin from them, since the fats, for the most part, become oxidized in the animal body, whereas in order to form cholesterin, they must undergo a process of deoxidation.

#### SEROLIN.

This body, which as yet has been very little studied, was discovered by Boudet,<sup>1</sup> in the solid residue of the serum of the blood.

At an ordinary temperature it appears in nacreous, glistening flocculi, which are very slightly soluble in cold, but dissolve pretty freely in hot alcohol, and in ether, and do not form an emulsion with water. This body has no reaction on vegetable colors, melts at  $+36^{\circ}$ , and apparently may be distilled with only partial change. The ammoniacal vapors and the very peculiar smell which it develops during distillation indicate that it contains nitrogen. It is not saponified by the alkalis.

Serolin is obtained by extracting, with hot alcohol, blood which has been dried, then boiling with water, and again dried. As the alcohol cools the serolin separates in flocculi.

#### CASTORIN.

This body occurs in *castoreum*; it crystallizes from boiling alcoholic solutions in small, four-sided needles, is pulverisable when dried, melts at a temperature exceeding  $100^{\circ}$ , is not saponifiable, and is converted by concentrated nitric acid into nitrogenous, crystallizable, *castoric-acid*.

#### AMBREIN.

Ambrein is the principal constituent of amber; it crystallizes in white needles grouped in stars or wart-like shapes, melts at  $37^{\circ}$ , cannot be saponified, and is converted by nitric acid into *ambreic acid*,  $C_{21}H_{18}N_6O_3$ , which is crystallizable, and forms yellow salts.

#### INOSTERIN.

Busch<sup>2</sup> has applied the term *Inosterin* to a non-saponifiable fatty

<sup>1</sup> Ann. de Chim. et de Phys. T. 52, p. 337.

<sup>2</sup> Müller's Arch. 1851, S. 358.

matter, which crystallizes in needles, fuses at a little above  $100^{\circ}$ , is soluble in cold and hot ether as well as in boiling alcohol, from which it evaporates in an amorphous shape. He discovered it in a uterine tumor; it probably also occurs in the adventitious products known as collonema and colloid.

## NON-NITROGENOUS NEUTRAL BODIES.

Most of the substances belonging to this class closely resemble one another in their empirical composition, and hence some of them have received the name of "carbo-hydrates;" for most of them contain hydrogen and oxygen in the same ratio as these elements are contained in water, so that if we suppose that they were combined into water, carbon would be the only remaining element of these bodies; indeed, even the number of atoms of carbon in them appears to be in accordance with a general rule, since in all the formulæ which as yet have been well established it is divisible by 6.

Considering their extremely analogous composition, it is naturally to be expected that these bodies should present many chemical properties in common with one another, various as their physical characters may be. They are so indifferent that it is only with few other bodies, and in these cases with considerable difficulty, that they can be made to combine, and then they enter into combination in multiple proportions, so that it is always difficult to determine their atomic weights with any degree of certainty. Almost the only physical properties which they have in common are deficiency in color and smell. They are all decomposed by heat, and yield acid products of distillation. By digestion with dilute mineral acids, they are for the most part converted into glucose or grape-sugar. When decomposed by nitric acid, they yield oxalic acid, saccharic acid, and mucic acid, and, perhaps, also, conjugated nitric acids. When treated with concentrated sulphuric acid these bodies become brown or black, and in addition to humin-like substances, form conjugated sulphuric acids.

The only substances of this group of any zoo-chemical importance are glucose or grape-sugar, milk-sugar, [inosite<sup>1</sup> or muscle-sugar,] and cellulose.



### *Chemical Relations.*

*Properties.*—Glucose, which is the name applied to grape-sugar by the French chemists, is identical with *diabetic sugar*, and crystallizes with 2 atoms of water in wart-like masses consisting of minute plates

<sup>1</sup> [Inosite or muscle-sugar has been discovered by Scherer since the original publication of this volume. Its formula is  $\text{C}_{12}\text{H}_{22}\text{O}_{11}$ . It will be noticed in a future part of this work.—G. E. D.]

arranged in a cauliflower form; these plates are rhombic and not square (as Saussure believed); when this substance separates rapidly from a solution, we may observe under the microscope that it occurs in irregularly striated, roundish masses, and not in plates; it is white, devoid of odor, and not so sweet as cane-sugar but sweeter than milk-sugar; it is only half as soluble in water as cane-sugar, but more soluble than milk-sugar; it is only slightly soluble in alcohol, and insoluble in ether; its aqueous solution turns the plane of polarization of a ray of light to the right, and is devoid of action on vegetable colors.

At a few degrees below  $100^{\circ}$  it begins to cake together, but it melts perfectly at  $100^{\circ}$  with the loss of its 2 atoms of water; at  $140^{\circ}$  it becomes converted into caramel, and develops a sweetish odor; at a higher temperature it becomes frothy, grows brown, develops a pungent vapor, and leaves a voluminous charcoal.

In contact with nitrogenous bodies, and especially with casein, it undergoes the lactic, and subsequently the butyric fermentation; with common yeast it passes into the state of vinous fermentation. Digested with concentrated nitric acid, it develops nitric oxide gas, and is converted into oxalic and saccharic acids; while chlorine gas converts it into hydrochloric and saccharic acids. When digested with dilute sulphuric acid, its solution does not so rapidly become brown as that of cane-sugar, and it is only on evaporation that we observe the formation of a blackish-brown residue; but its solution, when boiled with potash, very quickly assumes a fine brownish-red tint, and at the same time evolves a pungent, sweetish odor; it may be evaporated with lime-water without the development of any brown color, the lime and the sugar forming a syrupy compound with a bitter taste. On treating an aqueous solution of glucose with caustic potash, and then adding a salt of oxide of copper, no precipitate is formed, but the solution assumes a beautiful azure tint; after some time this gradually changes to a green color, and finally a red powder is deposited; if the fluid be boiled, it at once assumes a yellow tint, and suboxide of copper is separated as a yellow or yellowish-red powder. Glucose is distinguished by its property of forming a beautiful crystalline compound with chloride of sodium.

*Composition.*—According to the above formula, glucose consists of:

Carbon,	.	.	.	.	.	12 atoms,	.	.	.	40.000
Hydrogen,	.	.	.	.	.	12 "	.	.	.	6.666
Oxygen,	.	.	.	.	.	12 "	.	.	.	58.334
										100.000

Its atomic weight = 2250. (Peligot,<sup>1</sup> Erdmann, and Lehmann.<sup>2</sup>)

*Combinations.*—The compound of *glucose and potash*,  $2\text{KO} + \text{C}_{12}\text{H}_{12}\text{O}_{12}$ , is obtained by adding an alcoholic solution of caustic potash to an alcoholic solution of glucose; it occurs in the form of white flocculi, which, on exposure to the air, soon become tenacious and moist, and at length perfectly deliquescent; when dissolved in water they exhibit an alkaline reaction, and attract carbonic acid from the atmosphere.

The compound of *glucose and lime*,  $2\text{CaO} + \text{C}_{12}\text{H}_{12}\text{O}_{12}$ , is formed when

<sup>1</sup> Ann. de Ch. et de Phys. T. 66, p. 140, and Compt. rend. T. 6, p. 232.

<sup>2</sup> Journ. f. pr. Ch. Bd. 13, S. 113.

a solution of glucose is mixed with an excess of lime, and the filtered fluid treated with alcohol; it forms a white mass, which on exposure to the atmosphere attracts water and becomes transparent.

It is not easy to obtain a combination of *glucose with oxide of lead* in definite proportions; its aqueous solution takes up a large quantity of this oxide; an insoluble compound is obtained from glucose and a solution of acetate of lead treated with ammonia.

The combination of *glucose with chloride of sodium*,  $C_{12}H_{12}O_{12} + 2HIO + C_{12}H_{12}O_{12} \cdot NaCl$ , may be obtained by the direct mixture of the solutions of the two constituents and by spontaneous evaporation, in very large, colorless, four-sided, double pyramids. These crystals are hard, easily pulverizable, of 1.5441 specific gravity, transparent, unaffected by the atmosphere, of a saline and sweetish taste, soluble in 3.685 parts of cold water, and difficult of solution in alcohol. At  $100^{\circ}$  the powdered crystals begin to cake together, and lose 4% of water; when rapidly heated to  $120^{\circ}$  they melt in their water of crystallization, and begin to become brown at  $+160^{\circ}$ . The crystals contain 13.3% of chloride of sodium.

*Preparation.*—This sugar is not only, as is well known, widely diffused throughout the vegetable kingdom, but may be formed from other kinds of sugar and from carbo-hydrates (starch, wood-fibre, &c.) by digestion with dilute acids; hence it may be prepared in many different ways. On the large scale it is commonly obtained from starch; but all that concerns us here is its mode of preparation from diabetic urine. The following is the ordinary mode of proceeding. Diabetic urine is treated with basic acetate of lead, and the excess of lead removed from the filtered fluid by sulphuretted hydrogen; the fluid is then evaporated, and extracted with alcohol, from which the sugar crystallizes; but sugar thus obtained always contains acetates. In order to obtain the sugar I am in the habit of evaporating the urine to nearly the thickness of a syrup; provided it has not been too powerfully evaporated, the whole residue, after a variable time, becomes converted into a solid yellowish-white mass, which must be extracted with absolute alcohol and subsequently with hot spirit. The sugar dissolved in the latter is removed, after it has crystallized, by filtration, while the spirit is submitted to evaporation, and then treated with a little water in order to facilitate further crystallization. In this way we obtain the sugar in a state of greater purity than by the ordinary method.

In order to obtain diabetic sugar in a state of chemical purity, I prepared the chloride of sodium compound by saturating the aqueous solution of the alcoholic extract with chloride of sodium, and by repeated crystallization obtained it in perfectly transparent crystals, which I dissolved in water, and cautiously precipitated with sulphate of silver; the fluid freed by filtration from the chloride of silver was evaporated, and the sugar was obtained in a state of chemical purity by extraction with alcohol; in order to remove any traces of this fluid, it must be crystallized from distilled water.

*Tests.*—The methods of testing for sugar are not merely of importance in enabling the physician to establish his diagnosis in cases of Diabetes mellitus, but likewise in consequence of the physiological relations of sugar to the general metamorphosis of tissue. Many chemists

(amongst whom may be enumerated Golding Bird,<sup>1</sup> Gairdner,<sup>2</sup> Budge,<sup>3</sup> and myself),<sup>4</sup> have turned their attention to the most accurate methods of discovering sugar. There has been much discussion regarding Trommer's<sup>5</sup> admirable test for sugar; but if this test be not admitted, equal objections may be advanced against all the reagents employed in mineral chemistry; for these also require to be used with proper care and circumspection; the application of most of them demanding more precaution and skilful manipulation than this test. It may be regarded as infallible for the recognition of the presence of sugar in diabetic urine; although a person utterly ignorant of chemical reagents may also here fall into error. In true Diabetes mellitus, the urine is free from those substances which may interfere with the reaction on which this test is founded, or rather from the judgment we form regarding this reaction: diabetic urine presents this difference from other saccharine urine, that the former with sulphate of copper and potash gives the reaction almost as readily as a pure solution of grape-sugar would do, even when there is but little sugar present, whilst the more normal urine in which sugar is a mere incidental constituent, gives a less distinct reaction; and the latter moreover precipitates other substances with the suboxide of copper, by which the color of the precipitate is considerably modified.

The question here arises—what precautionary measures ought to be observed in the application of Trommer's test?

The fluid to be examined is treated with *caustic potash*, and filtered if necessary, that is to say, if there be too great a precipitate; an excess of caustic potash is productive of no harm, as it should be present in more than sufficient quantity to decompose the *sulphate of copper*; the latter, which must be added gradually, and in a diluted state, usually gives rise to a precipitate, which disappears when the fluid is stirred; as the quantity of the oxide of copper which is soluble is proportional to the quantity of sugar which is present, very little sulphate of copper must be added at a time, if we suspect that only a little sugar is present in the fluid. On allowing the azure solution thus obtained to stand for some time, there is usually formed a more pure red or yellow powder than the precipitate which is at once thrown down on boiling the fluid. Moreover, very prolonged heating is improper, for there are several substances which by prolonged boiling separate suboxide of copper from alkaline solutions of oxide of copper; amongst them we may especially name the albuminous substances, which with oxide of copper and potash yield very beautiful azure-blue, or somewhat violet solutions, and by very prolonged boiling, separate a little suboxide of copper, although without the aid of heat they have not this property.

If a specimen of urine contain very little sugar, or if we are searching for sugar in some other fluid, it is advisable to extract the solid residue with alcohol, to dissolve the alcoholic extract in water, and to apply the potash and sulphate of copper to this solution. By proceeding in this manner we usually obtain the reaction in its most distinct manner.

<sup>1</sup> Monthly Journal of Medical Science, Vol. 4, p. 423, [and "Urinary Deposits," 3d Ed. p. 352.—G. E. D.]

<sup>2</sup> Ibid. p. 564.

<sup>4</sup> Schmidt's Jahrb. Bd. 45, S. 6–10.

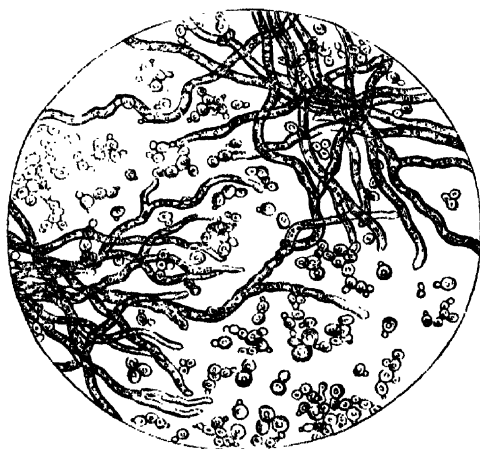
<sup>3</sup> Arch. f. physiolog. Heilk. Bd. 3, S. 385.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 39, S. 860.

If, however, we are seeking for very small quantities of sugar, as for instance in chyle, blood, or in the egg, we must neutralize the aqueous fluid, previously to its evaporation, with dilute acetic acid, in consequence of the solubility of albuminate of soda or of casein in alcohol, thus preventing any albuminous body from remaining in solution. If the reaction do not properly manifest itself in the alcoholic extract thus obtained, or if we would carry the investigation further, we must precipitate the sugar from the alcoholic solution by an alcoholic solution of potash, dissolve the compound of sugar and potash in water, and now apply the sulphate of copper; if only a trace of sugar be present, we obtain a most distinct and beautiful reaction.

The *fermentation-test* has been much extolled as a means of discovering sugar; but independently of the circumstance that the process is very tedious, it yields, to an inexperienced experimentalist and observer, far less certain results than Trommer's test. On adding yeast to a fluid, the phenomena of fermentation are simply dependent on the development of bubbles of carbonic acid; if this development of gas from a fluid, as, for instance, from diabetic urine, be not very active after the addition of yeast, we must not draw any conclusions regarding the presence of sugar, for yeast promotes the decomposition of the animal fluids—a process which is often accompanied with the development of a little gas. If, however, no yeast be added to the urine, but we wait for spontaneous fermentation, as has also been recommended, the development of carbonic acid proceeds very slowly, unless an extremely large quantity of sugar be present; moreover, in this case, there is this additional difficulty in observing the formation of the gas, that the sugar for the most part undergoes the lactic and not the vinous fermentation. As the detection of the alcohol, which is formed during this process, is by no means easy, attention has been drawn to the formation of the yeast-fungus (*Torula cerevisiæ*) as a characteristic indication of

Fig. 17.

*Torula cerevisiæ*, or Yeast-fungus.

vinous fermentation. For those who are accustomed to the use of the microscope, and are well acquainted with the appearance of the *Torula*, this

is unquestionably an easy and certain test; but it must be borne in mind, that when normal urine has been allowed to stand for a long time, especially at a high temperature, fungi of a precisely similar shape are formed in it, probably, for the most part, from the mucus. These fungi, which are by no means dependent on the decomposition of sugar, may exist in urine still preserving a decidedly acid reaction, although they more frequently occur in neutral urine; the individual cells, which like the yeast-cells (*Torula cerevisia*), contain distinct nuclei, are mostly about one-half (in diameter) smaller than the true yeast-cells; but independently of the circumstance that under the microscope apparent magnitudes afford a very relative criterion, the yeast-cells which are first and spontaneously formed, are always much smaller than those which are subsequently produced by gemmation from previously formed yeast-fungi.

A very good means of discovering sugar, and of determining its quantity with considerable accuracy in a clear solution, is afforded by *Biot and Seil's polarizing apparatus*; its expense will, however, always stand in the way of its general application.

We have already shown (p. 118) that *Pettenkofer's test* is not available for the detection of sugar.

All other tests which were formerly employed for the discovery of sugar (evaporation with hydrochloric or sulphuric acid, treatment with chromic acid, boiling with caustic potash, &c.) are open to so many sources of fallacy, as compared with the methods we have already indicated, that we may pass them over in silence.

The test proposed by Maumene<sup>1</sup> not only gives precisely the same reaction with glucose and the other true sugars, but also with other carbohydrates; for on heating all such substances to 100°, after the application of chlorine or metallic chlorides, they are converted into glistening black masses; this happens not only with sugar, but with woody fibre, hemp, linen, cotton, paper, starch, &c. Hence it is easy to see that this method is open to many fallacies, from the accidental presence of shreds of paper, dust, &c. If, however, we had to deal with pure substances, we might certainly recognize very small quantities of sugar, if, in accordance with Maumene's directions, we moisten, with the fluid to be investigated and then heat to 100° a pure woollen tissue (merino) which had been previously saturated with a solution of chloride of tin and afterwards dried. A glistening black patch is formed at the spot that had been moistened.

Trommer's test may also be successfully employed in the *quantitative determination* of the sugar in diabetic urine; Bareswil,<sup>2</sup> Falck,<sup>3</sup> and Scharlau,<sup>4</sup> have recommended different methods of applying it with this view; the most generally applicable one, however, is that of Fehling.<sup>5</sup> As a test he uses a solution of 40 grammes of crystallized sulphate of copper in 160 grammes of water; this is mixed with a concentrated solution of 160 grammes of tartrate of potash and 560 grammes of a solution of caustic-soda (specific gravity = 1.12), and water is then added

<sup>1</sup> Compt. rend. T. 30, pp. 314 et 447.

<sup>2</sup> Journ. de Pharm. T. 6, p. 301.

<sup>3</sup> Oesterlen's Jahrb. f. pr. Heilk. Bd. 1, S. 509.

<sup>4</sup> Die Zuckerharnruhr. Berlin, 1846.

<sup>5</sup> Arch. f. phys. Heilk. Bd. 7, S. 64-73.



till the volume of the fluid at  $+15^{\circ}$  amounts to 1 litre. 11.5c.c. of a saccharine solution containing 5 grammes of dry sugar ( $=C_{12}H_{12}O_{12}$ ) in a litre, are necessary to cause the perfect reduction of the oxide to the suboxide of copper in 10c.c. of the test-fluid. Hence it follows that 100 parts of oxide of copper are completely reduced to the state of suboxide by 45.25 parts of sugar, or 10 atoms of oxide of copper by 1 atom of sugar. In order to determine with the greatest certainty the weight of the sugar from the volumetric measurement, and to render the errors of observation as small as possible, Fehling recommends that the urine to be examined for sugar should be diluted with water to 10 or 20 times its volume, that is to say, that 50 grammes of urine should be treated with 450 or 950 of water: 10c.c. of the test-fluid are then to be diluted with about 40c.c. of water, boiled, and so much of the diluted urine (which must be kept in a burette or graduated tube in order that we may be able to estimate the quantity used) added to it, as to effect as nearly as possible the complete decomposition of the sugar and of the oxide of copper. If any undecomposed oxide of copper be contained in the fluid after the removal of the suboxide by filtration, it may be recognized by the blue tint, and by its reaction with sulphuretted hydrogen: if, on the other hand, too much urine be added, the filtered fluid appears yellow from the action of the caustic alkali on the sugar. The point of thorough mutual decomposition is easily attained by a few repetitions of the experiment. As 10 c.c. of the test-fluid require 0.0577 of a gramme of sugar for the reduction of the oxide of copper contained in them, there must be exactly that amount of sugar in the quantity of urine used in the experiment, and hence the proportion of sugar in any given quantity of urine may be easily calculated. Moreover, this method must be employed with considerable circumspection in order to yield accurate results: for if the fluid which is being examined contains other organic substances, which can either combine with the alkali of the test-fluid, or can decompose oxide of copper even to a slight degree, either the alcohol-extract of the fluid, or the sugar-and-potash-compound, must be first exhibited before the test can be applied. A second objection to this procedure is, that we cannot keep the test-fluid for any length of time, and that we, consequently, ought to prepare a fresh quantity for each determination of sugar. In the course of time the alkali exerts a modifying action on the tartaric acid, so as to give it the property of reducing the oxide of copper with the co-operation of heat, and, indeed, even in the cold. If we have boiled the freshly prepared solution with the greatest care, and have convinced ourselves that no suboxide is precipitated, we still find that after a week a little of the suboxide is separated on boiling, and after it has stood for a longer time, a similar decomposition ensues, even at an ordinary temperature. This method is, however, by no means to be rejected for this deficiency: but at the same time we must not overlook it.

Those who are not in the habit of using French weights and measures may prepare Fehling's test solution as follows:—Dissolve 69 grains of crystallized sulphate of copper in five times their weight of distilled water, and add to it, first, a concentrated solution of 268 grains of tartrate of potash, and then a solution of 80 grains of hydrate of soda in

one ounce of distilled water. Put the solution into an alkalimeter tube, and add distilled water so as to make 1000 grain-measures of the liquor. This solution will be nearly double the strength of that made according to the above directions, and every 100 grain-measures of it will be equivalent to 1 grain of grape sugar. [G. E. D.]

By *Soleil's polarizing apparatus* the quantity of sugar may be determined with more rapidity than by the preceding method, and with equal accuracy. Many precautions are, however, requisite in its application, as has been especially shown by Dubrunfaut,<sup>1</sup> Clerget,<sup>2</sup> and Lespiaux.<sup>3</sup> We refer, therefore, to their communications on this subject; especially as Soleil's apparatus, in so far as its application to saccharine urine is concerned, is still deficient in many respects.

*Fermentation* was formerly employed to determine the quantity of sugar in fluids, the carbonic acid being determined, and the quantity of sugar calculated from it. This mode of determination is deficient in accuracy, in the first place, because all animal fluids, and especially the urine, contain free carbonic acid, and, secondly, because other constituents of the urine are simultaneously decomposed during the process of fermentation, and also yield carbonic acid. This method serves, however, very well for approximate and comparative determinations, if we allow a weighed quantity of diabetic urine to ferment at 37° in Fresenius's<sup>4</sup> alkalimetical apparatus, and, as in alkalimetical processes, determine the carbonic acid by the loss of weight.

This is the best means of determining the amount of sugar for ordinary medical purposes, Fehling's method being applicable rather to technology than to clinical medicine. If the apparatus be allowed to stand for about 48 hours at the above-mentioned temperature, all the sugar will have been converted into spirit; if we even omit to remove the carbonic acid by drawing a little air through the apparatus, previously to weighing, we shall still obtain results at all events sufficiently accurate for purposes of comparison.

#### *Physiological Relations.*

*Occurrence.*—In a normal condition of the system this form of sugar may always be recognized in the *primæ viæ*, especially in the contents of the small intestine after the use of vegetable, that is to say, of amylaceous and saccharine food. We shall subsequently see, when treating of digestion, that it is principally through the influence of the pancreatic juice that starch is gradually converted, in the intestinal canal, into sugar. It is only in small quantity that it exists in the contents of the small intestine, partly because the change effected in the starch proceeds very gradually, and partly because the sugar which is formed is very rapidly absorbed.

Trommer<sup>5</sup> was the first who detected traces of sugar in the *chyle*; I have several times most distinctly recognized the presence of sugar in the chyle of horses which, a few hours before they were killed, had taken either pure starch or strongly amylaceous food.

Sugar undoubtedly exists in the *blood*; Magendie<sup>6</sup> asserts that he

<sup>1</sup> Ann. d. Chim. et de Phys. 3 Sér. T. 18, p. 101.

<sup>2</sup> Compt. rend. T. 22, p. 200, and pp. 256-260.

<sup>3</sup> Ibid. T. 26, p. 306.

<sup>4</sup> Neue Verfahrungsweisen zur Prüfung der Soda, &c. Heidelb 1843.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 39, S. 360.

<sup>6</sup> Compt. rend. T. 30, p. 191-192.

found considerable quantities of sugar, together with dextrin, in the blood of a dog, which for several days had been fed solely on boiled potatoes. C. Schmidt<sup>1</sup> has subsequently shown that it is a normal constituent of the blood of oxen, dogs, cats, and men; and I<sup>2</sup> have since found (almost simultaneously with Cl. Bernard<sup>3</sup>) that the portal blood contains no sugar, or only traces of that substance, while the blood of the hepatic veins is very rich in sugar. Bernard has satisfied himself that the sugar in the vessels near the heart gradually diminishes, and that only very little can be found in the arterial blood.

In a normal state it is probable that no sugar finds its way into the urine; at least after living for two days solely on fat and sugar, I was as unsuccessful in the search for sugar in my urine, as Magendie had been in the case of the dog in whose blood he found sugar.

It is only seldom that we meet with saccharine urine in *other diseases* than diabetes. Prout has sometimes found sugar in the urine of "gouty and dyspeptic persons," and Budge<sup>4</sup> in "abdominal affections and hypochondriasis." I<sup>5</sup> once met with sugar in the urine of a puerperal woman, in whom, on the fifth day after delivery, the secretion of milk was suddenly suspended. I was led to the discovery that it contained sugar by observing the formation of yeast-cells in it; the sugar only continued in the urine of this woman for four days. I have recently found sugar in the urine of a man suffering from a very well-marked attack of arthritis.

Alvaro Reynoso<sup>6</sup> has recently believed that he has found sugar in the urine in various bodily conditions, especially in cases of epilepsy and hysteria; he further believes that sugar is constantly to be found in the urine after narcotism has been induced by the inhalation of ether, also in pulmonary affections, and after the employment of the so-called hyposthenic agents, as metallic salts, sulphate of quinine, narcotic drugs, &c.; Uhle, who has had opportunities of most carefully examining the urine in all these conditions (for the most part under my own direct superintendence), has never been able to confirm any one of Reynoso's observations.

Bernard<sup>7</sup> found sugar in considerable quantity in the urine of the foetus of the cow between the fifth and seventh months, and in that of the sheep at two months.

The same observer also found sugar in the fluids of the amnios and allantois of the calf, lamb, and pig. In a seven months' foetal calf, sugar was found in the urine; but it no longer existed in the above-mentioned fluids.

Although I have myself<sup>8</sup> once found sugar in the *saliva*, in a case of acute rheumatism, in which spontaneous salivation ensued, and this secretion was discharged in great abundance, I cannot venture to conclude from this isolated instance that sugar ever exists in the saliva of non-

<sup>1</sup> Charakteristik der Cholera u. s. w. 1850, S. 161-168.

<sup>2</sup> Ber. d. Gesellsch. d. Wiss. zu Leipzig. 1850, Bd. 3, S. 139-141.

<sup>3</sup> Arch. gén. de Méd. T. 18, p. 303.

<sup>4</sup> Arch. f. physiol. Heilk. Bd. 3, S. 413.

<sup>5</sup> Jahresh. d. physiol. Ch. 1844, S. 27.

<sup>6</sup> Compt. rend. T. 33, p. 410-416, p. 521, and T. 34, p. 18.

<sup>7</sup> Ibid. Vol. 30, p. 317.

<sup>8</sup> Jahresh. d. Physiol. Ch. 1844, S. 20.

diabetic persons, since in this case it is possible that the sugar might in some way have accidentally got into the vessel containing the saliva. So many heterogeneous substances find their way into the saliva, as we shall subsequently see, that there is nothing extravagant in the assumption that sugar may sometimes occur in morbid saliva. Wright places a sweet saliva amongst his numerous varieties; unfortunately, however, he did not proceed to ascertain whether the sweetness of this saliva was dependent on the presence of sugar, or whether it was a mere subjective sensation of the patient.

F. L. Winkler<sup>1</sup> found 8 grains of sugar in two softly-boiled eggs, which had been sat upon for some time, and whose white had a singularly sweet taste. I have recently convinced myself that small quantities of sugar are constantly present both in the *yolk* and in the *white* of fresh eggs.

I may remark that I experimented upon 30 eggs, in order to obtain evidence of the existence of small quantities of sugar. I have repeatedly, and with much care, repeated Winkler's experiment, in which he found so large a quantity of sugar (milk-sugar) in incubated eggs, but, I cannot confirm his statement. I examined eggs that had been sat upon for 3, 7, and 15 days.

Bernard and Barreswil<sup>2</sup> have found sugar in the tissue of the liver even of animals that do not subsist on saccharine or amylaceous food.<sup>3</sup> Bernard<sup>4</sup> has subsequently taken up this subject much more fully, while the fact that sugar exists in the hepatic tissue has been thoroughly confirmed by Frerichs,<sup>5</sup> Baumert,<sup>6</sup> and myself. The amount of sugar in the liver is much more considerable in many mammals and birds than in reptiles, while in the liver of fishes there appears to be no sugar. At all events, Bernard found no trace of sugar in the liver either of the eel or of the skate. In many diseases the sugar disappears from the liver.

Sugar has been sought for in all the fluids in cases of *Diabetes*, and has been so generally found that it is unnecessary to quote authorities on the subject. It has been found not merely in the urine, blood, and all serous fluids, but also in the saliva, in vomited matters, in the solid excrements, and even in the sweat.

In a person suffering from well-developed diabetes, and who, at the same time perspired very freely (a combination not often observed), it was only in the sweat that I failed to detect sugar.

In examining the body of a person who died from diabetes, Bernard only failed in detecting sugar in the following organs, viz., the brain and spinal cord, the pancreas and the spleen.

*Origin.*—The origin of the small quantities of glucose which normally occur in the animal fluids, is so obvious, as hardly to require notice. I will here only remark, that little or nothing in the way of conclusion can be deduced in reference to the metamorphosis of starch or

<sup>1</sup> Buchn. Report, Bd. 42, S. 46.

<sup>2</sup> Compt. rend. T. 27, p. 514.

<sup>3</sup> [Experiments conducted in the Giessen laboratory have confirmed this statement, both in reference to the livers of graminivorous and carnivorous animals. See Liebig and Kopp's Annual Report, &c., for 1847-8, Vol. 2, p. 175, note 6.—G. E. D.]

<sup>4</sup> Compt. rend. T. 31, p. 572-574.

<sup>5</sup> Handwörterbuch d. Physiologie. Bd. 3, Abt. 1, S. 831.

<sup>6</sup> Journ. f. pr. Ch. Bd. 54, S. 857-863.

dextrin within the animal organism from experimental attempts to convert starch into sugar by means of saliva, the serum of the blood, renal tissue, &c.; for any other nitrogenous substance acts just as efficiently, if it be digested for a sufficiently long time with water and starch-paste, in converting a portion of the latter into sugar. The actual substance, which, in all probability, effects the conversion of starch into sugar, is, as we have already mentioned, the pancreatic juice. Magendie's experiment,<sup>1</sup> in which starch was converted into sugar in the circulating blood of a living animal, proves little in relation to the physiological process, since starch does not normally pass into the blood. We shall enter more fully into the consideration of the digestion of starch, and of the experiments bearing on this point which have been instituted by Bouchardat and Sandras, Jacobowitsch, Strahl, and others, in a future part of the work.

But whence originates the enormous quantity of sugar which, in *diabetes*, is often discharged with the urine? While no one can doubt that it is, for the most part, at all events, derived from vegetable food, it is still a contested question whether the nitrogenous constituents of the animal body may not also contribute to the formation of this substance. Many have assumed it as beyond all question (Budge<sup>2</sup>), that in *diabetes*, sugar is formed from protein, but, on examining the grounds on which such a view is based, we find that the facts adduced in support of them are of a very doubtful character. In the first place, it has been alleged that diabetic patients, living on a highly nitrogenous diet, discharge far more sugar than could be formed from the sugar-yielding, non-nitrogenous substances, which have constituted a portion of their food; but unfortunately no accurate observations on this point, based on numerical results, have been brought forward; for, although Pfeuffer and Löwig<sup>3</sup> have instituted one experiment of the kind, it led to no result. Moreover, we are still so ignorant regarding the internal constitution of albuminous and gelatinous substances, that we can adduce no chemical grounds in support of such an assumption. Berzelius,<sup>4</sup> founding his hypothesis on the fact that protein, like sugar, when treated with hydrochloric acid, yields formic and humic acids, and, with nitric acid, yields oxalic and saccharic acids (which, however, has not been decisively proved), indicates the possibility that protein (like amygdalin, salicin, &c.), may contain sugar, and that a portion of the diabetic sugar may, therefore, proceed from the albuminous substances. The supposition is, also, by no means at variance with the admirable investigations of Guckelberger on the products of decomposition of nitrogenous animal tissues by sulphuric acid and chromate of potash; since by this means of oxidation, aldehyde<sup>5</sup> is developed from these nitrogenous matters, just as it is produced from milk-sugar when similarly treated. These facts, however, simply indicate the possibility that sugar may be formed from the protein-compounds; they do not prove that it is so formed; Liebig<sup>6</sup> merely regards it as "conceivable" that in the metamorphosis of tissue, sugar may be produced from gelatinous substances.

<sup>1</sup> Compt. rend. T. 30, pp. 189-192.

<sup>2</sup> Arch. f. physiol. Heilk. Bd. 3, S. 391.

<sup>3</sup> Jahresber. Bd. 19, S. 655.

<sup>6</sup> Geiger's Pharm. Bd. 1, S. 796.

<sup>5</sup> Zeitsch. f. rat. Med. Bd. 1, S. 451.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 64, S. 93.

Notwithstanding the numerous hypotheses that have been advanced by physicians regarding the reason why, in diabetes, the sugar does not undergo the ordinary change in the organism, we are still utterly ignorant on this point. As we shall return to this subject in the second volume, in our observations on "the urine," it will suffice if we here mention the following facts, which may subsequently influence our judgment in reference to this matter.

I<sup>1</sup> injected two drachms of cane-sugar dissolved in water, into the veins of a dog; the dog, who had lost very little blood during the operation, drank a great deal, and discharged a large quantity of sweet-tasting urine, which contained *unchanged* cane-sugar; and Kersting<sup>2</sup> arrived at a similar result with other kinds of sugar. Bernard<sup>3</sup> injected a solution of cane-sugar into the veins of a dog and a rabbit; the urine of the animals remained acid, and contained unchanged cane-sugar; but, on repeating the experiment on another dog and rabbit with a solution of glucose, he failed to detect this substance in the urine, which had now become alkaline.

[The admirable experiments and observations of Dr. Percy on this subject are apparently unknown to Professor Lehmann. See the "Medical Gazette," Vol. 32, pp. 19, 591, and 640.—G. E. D.]

If we were to attempt to draw any conclusion from these few experiments, it would be that in diabetes the glucose formed from the vegetable substances in the intestine is not, as in the normal state, metamorphosed in the blood. We have been in the habit of referring the alkaline reaction of the urine in gramnivorous animals, to the decomposition of the salts formed by organic acids and the alkalis into carbonates, but from Bernard's experiment, it would appear as if the alkalinescence were dependent on other relations connected with the nature of the vegetable food: at all events, I found that, when for two entire days I took nothing but sugar, fat, and starch, and consequently food devoid of nitrogen and free from alkalis, my urine had an extremely weak acid reaction.

More accurate investigations, or a more detailed account of his mode of procedure are requisite, before we can form an opinion regarding certain experiments performed by Bernard,<sup>4</sup> or can attempt to explain them on physiological grounds. He maintains, that he has found sugar in the urine and the blood, after irritating one definite spot in the base of the fourth ventricle of the brain. This experiment, if it should receive further confirmation, will apparently strengthen Scharlau's hypothesis that diabetes is essentially a disease of the spinal cord, unless Bernard associates it with the function of the pneumogastric nerves; for when they have been divided he has also found sugar.

*Uses.*—Since glucose, which, as we have already seen, is principally formed in the intestinal canal from the starch of the vegetable food, appears, from the results of all physiological inquiries, to be a true element of nutrition (see "Nutrition"), the question that remains to be considered is—how it is applied, or what is its use in the animal organ-

<sup>1</sup> Jahresber. d. phys. Ch. 1844, S. 47.

<sup>2</sup> Diss. inaug. med. Lips. 1844.

<sup>3</sup> Compt. rend. T. 22, pp. 534-537.

<sup>4</sup> Ibid. T. 28, p. 393, and Arch. gén. de Méd. 4 Sér. T. 18.

ism? It belongs, according to Liebig, to the food for the respiration; and if we regard it purely in this light, its object is easily understood; it undergoes a process of combustion by combining with the inspired oxygen, its final products being water and carbonic acid, and tends to support the animal heat, if we regard this as an independent process. If, however, we entirely concur in this view, we have still to inquire whether the sugar does not previously undergo other changes and serve other objects, before it yields carbonic acid and water as the final products of its combustion.

It must excite our surprise that in diabetes, where, in reference to the respiration, the saccharine and amylaceous elements of food appear to be entirely lost, the respiration and the animal heat are so well supported; for, although pulmonary tuberculosis is a frequent complication of diabetes, this is by no means invariably the case; and it may occur without any affection of the lungs. It certainly seems very remarkable that such a mass of the respiratory food can be lost without inducing any symptom of a disturbed respiration or of a diminished animal heat.

We have already referred (p. 230) to the hypothesis of the conversion of sugar in the intestinal canal into *fat*, and shown that it is unsupported by facts; but we do not deny that in some part of the animal body (at least under certain relations) sugar may be metamorphosed into fat. Moreover, we are still so ignorant regarding the different changes which the sugar undergoes in the blood, that, to a certain degree, we must content ourselves with the consideration of questions that may lead us on the true path of inquiry. We have already pointed out the probability that the *lactic acid* occurring in the animal body, is formed from sugar (p. 101); under special relations *butyric acid* may also be produced from it (p. 64). The alkalescence of the urine observed by Bernard after the injection of glucose, would almost seem to indicate that the sugar in the blood is converted into an acid, which, combining with the alkali of the blood, yields carbonated alkali as a product of combustion, which passes into the urine and renders it alkaline. This experiment undoubtedly shows that the principal metamorphosis of the sugar occurs primarily in the blood, and not in the intestinal canal.

That the sugar undergoes *vinous fermentation* in the intestinal canal is a view that is now entirely rejected; for the yeast-corpuscles which we sometimes find in the contents of the intestines, and which might lead to the suspicion of such a fermentation, may take their origin from the food, as, for instance, from bread.

Does the sugar take any part in the formation of bile? We have already attempted (see p. 120 and p. 240) to show the probability that the bile is in part formed from fat, and that cholic acid should be regarded as conjugated oleic acid with the adjunct  $C_{12}H_6O_6$ . Can this adjunct take its origin from the sugar?

Those who assume that sugar exists preformed in nitrogenous animal substances, whether gelatinous or albuminous (as for instance it does in amygdalin), need feel no difficulty in believing that in the animal body protein is primarily formed from nitrogenous matters and sugar. In the case of chitin, however (to which further reference will be made in a

future page), we appear rather to have a combination of vegetable fibre with a nitrogenous substance.

We can hardly entertain a doubt that in the female mammalia the milk-sugar is derived from the glucose, but by what means this change is accomplished is a point on which we are entirely ignorant.

### MILK-SUGAR.— $C_{10}H_{18}O_8$ .

#### *Chemical Relations.*

*Properties.*—This substance forms white, opaque, overlying prisms or rhombohedra containing 2 atoms of water, is hard, crunches between the teeth, has a very faintly sweet, almost floury taste, is devoid of smell, dissolves slowly in cold but more readily in hot water, and is insoluble in absolute alcohol and ether; the aqueous solution, which moreover turns the plane of polarization of a ray of light to the right, may be evaporated to a very considerable extent without any separation of the sugar in a crystalline form.

When heated, milk-sugar melts, swells up, develops a sweetish pungent odor, and burns with a flame.

Digested with dilute sulphuric or hydrochloric acid, or with acetic or citric acid, it becomes converted into glucose; it absorbs large quantities of chlorine, hydrochloric acid, and ammoniacal gases. Nitric acid converts it into mucic acid with a little oxalic, saccharic, and carbonic acid; with sulphuric acid and bichromate of potash it yields not only formic acid but aldehyde.

In contact with the caustic fixed alkalies it becomes converted at  $225^{\circ}$  into oxalic acid; boiled with dilute alkalies or oxide of lead and water it becomes yellow or brown; at  $50^{\circ}$  it yields several compounds with oxide of lead. It reacts with sulphate of copper and potash exactly in the same manner as glucose. It was for a long time classed among the non-fermentable kinds of sugar, till Schill<sup>1</sup> and Hess<sup>2</sup> almost simultaneously remarked that milk-sugar only required a longer period in order to pass into a state of vinous fermentation under the influence of yeast, sour dough, gelatin, or albumen. H. Rose<sup>3</sup> has confirmed Schill's observations, that the formation of dextrin must precede the vinous fermentation of the milk-sugar, as indeed Payen had previously observed in reference to the sugar of the dahlia, and Rose in reference to cane-sugar. Like the other varieties of sugar, it can undergo lactic and butyric fermentation when the necessary ferments are added to it.

*Composition.*—In its crystalline state milk-sugar has exactly the same empirical formula as anhydrous glucose, so that it therefore contains equal equivalents of carbon, hydrogen, and oxygen. But as, when warmed, it loses 11.9% of water, that is to say, 1 atom of water to 5 atoms of carbon, its formula must either  $=C_5H_4O_4$  or a multiple of it. As milk-sugar cannot be combined with any body in a definite proportion, its true atomic weight is unknown. Its relation to nitric acid, with

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 31, S. 152.

<sup>2</sup> Pogg. Ann. Bd. 31, S. 194.

<sup>3</sup> Ibid. Bd. 52, S. 298.



which, as we have already mentioned, it yields mucic acid, shows that its constitution must in some respects be different from that of the other fermentable sugars.

*Preparation.*—Milk-sugar is obtained on the large scale by evaporating whey, and allowing the concentrated fluid to stand for a long time in a cool place. The crystalline incrustations which are then formed are purified by recrystallization. Simon recommends that the milk should be evaporated to  $\frac{1}{8}$ th of its volume, and that the casein should be precipitated by alcohol; the filtered fluid must be then further evaporated and treated with strong alcohol; the milk-sugar, which is precipitated with the water-extract, is then rinsed with a little water, dissolved in pure water, and left to spontaneous evaporation.

According to Haidlen<sup>1</sup> the milk should be boiled with  $\frac{1}{8}$ th its weight of pulverized gypsum, which coagulates the casein; the filtered fluid is then to be evaporated to dryness, and after the fat has been removed by ether, the milk-sugar may be extracted from the residue by boiling alcohol, which yields it in a state of perfect purity.

*Tests.*—If it be shown by Trommer's test that some kind of sugar is contained in the alcoholic extract of an animal fluid, we may readily distinguish milk-sugar from other kinds of sugar (if we have a sufficient amount of material to examine), by its difficult solubility in alcohol, by the slowness with which it ferments in the presence of yeast, and by its property of yielding the insoluble mucic acid when boiled with nitric acid. It may be estimated *quantitatively* with tolerable accuracy by Haidlen's method given above; but when extreme accuracy is required we must use Barreswil's or Fehling's test-fluid, in the manner described for grape-sugar (see p. 254); Poggiale has in this way determined the sugar in cows' milk by a test-fluid (consisting of 10 parts of crystallized sulphate of copper, 10 of bitartrate of potash, 30 of caustic potash, and 200 of distilled water), but his results were obviously in excess; for although he attempted to remove the casein previously with acetic acid, a portion of this substance must have remained in solution and co-operated with the sugar in decomposing the oxide of copper. A better method of proceeding is to remove the casein by boiling the milk with sulphate of magnesia or chloride of calcium, precipitating any excess of the earth from the filtered fluid with potash, and then applying Fehling's test-fluid; while perhaps the best is to proceed according to Haidlen's plan, and then to apply Fehling's method to determine the quantity of milk-sugar in the alcoholic extract.

### *Physiological Relations.*

*Occurrence.*—This substance appears to be an integral constituent of the milk of all mammalia. In woman's milk its amount ranges from 3·2 to 6·24% (Fr. Simon,<sup>2</sup> Haidlen,<sup>3</sup> Clémme<sup>4</sup>); in cows' milk it is stated to average from 3·4 to 4·3%; but by an improved method of analysis I have always found rather a larger amount of sugar in good cows' milk; but

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 45, S. 275.

<sup>2</sup> Frauenmilch. S. 35.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 45, S. 275.

<sup>4</sup> Handwörterbuch. d. Phys. Bd. 2, S. 464.

the average (—5·28%) assumed by Poggiale<sup>1</sup> is obviously too high; in that of the ass it constitutes 4·5%; in that of the mare, 8·7%, in that of the goat, 4·4%, and in that of the sheep 4·2%; indeed, it was even found in the milk of a he-goat (Schlossberger).<sup>2</sup> Dumas<sup>3</sup> thought that he had ascertained that the milk of bitches restricted entirely to an animal diet contained no milk-sugar, but it was subsequently ascertained by Bensch<sup>4</sup> that even then traces of milk-sugar were present; its quantity is however perceptibly increased under the use of a vegetable diet.

In the colostrum Simon found 7%, and in the milk six days after delivery only 6·24% of milk-sugar; his investigations show that it diminishes according to the length of time after delivery at which it is secreted, and that neither an *abundant* nor an *insufficient diet* influences its quantity, although differences in the food considerably affect the amount of butter. The observations of Donne,<sup>5</sup> Meggenhofen,<sup>6</sup> and Simon<sup>7</sup> concur in showing that *diseases*, especially syphilis, do not modify the amount of sugar in the milk.

Milk-sugar has been sought for in the *blood* by Mitscherlich, and Tiedemann and Gmelin, but hitherto without success. Though Guillot and Leblanc<sup>8</sup> believe that they have discovered milk-sugar in the blood of milch-cows.

[Braconnot<sup>9</sup> believes that he has demonstrated that milk-sugar exists in the cotyledons of the seeds of vegetables.—G. E. D.]

*Origin*.—The positive experiments of Dumas and Bensch, which prove that the amount of milk-sugar increases during a vegetable diet, give great probability to the opinion that this substance is principally formed from glucose or from the starch of the food; but notwithstanding the apparently affirmative observations of Bensch, the question whether it may not also be formed from nitrogenous matters, must for the present remain undecided. Where and by what means this conversion of glucose within the organism occurs, are subjects of which we are entirely ignorant.

*Uses*.—No doubt can be entertained that the milk-sugar which the infant at the breast receives in its food serves the same purposes in the economy that starch and other carbo-hydrates serve in the more matured organism.

#### INOSITE.— $C_{12}H_{12}O_{12}$ .

*Properties*.—This variety of sugar crystallizes with four atoms of water in colorless clino-rectangular prisms, which effloresce on exposure to the air, and lose all their water of crystallization at 100° or in vacuo; it has a sweet taste, dissolves readily in water, slightly in strong spirit, and is insoluble in alcohol and ether; it crystallizes from a boiling spirituous solution on cooling in glistening tablets somewhat like cholesterin; at a temperature exceeding 210° it fuses

<sup>1</sup> Compt. rend. T. 28, pp. 505–7.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 51, S. 431.

<sup>3</sup> Compt. rend. T. 21, pp. 708–717.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 221–227.

<sup>5</sup> Du lait et en particulier de celui des nourrices. Paris, 1836.

<sup>6</sup> Diss. inaug. sist. indagatiōnem lactis muliebris. Francof. a. M. 1816.

<sup>7</sup> Die Frauenmilch. Berlin, 1838.

<sup>8</sup> Compt. rend. T. 31, p. 585.

<sup>9</sup> Ann. de Chim. et de Phys. 4 Ser. T. 27, p. 399.

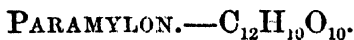
into a clear fluid, and at a still higher temperature it undergoes decomposition in the same manner as sugar. It undergoes no change when evaporated with hydrochloric acid, or when boiled with caustic potash. It forms a blue solution with sulphate of copper and potash; but there is no separation of suboxide of copper either on prolonged standing or on boiling; it does not undergo the vinous fermentation with yeast, but in the presence of casein or flesh it enters into lactic and butyric fermentation.

*Composition.*—Scherer,<sup>1</sup> the discoverer of this substance, has found that in the anhydrous state it is perfectly isomeric with anhydrous grape-sugar.

*Preparation.*—If we treat the muscular juice of the heart of the ox in the same manner as in the preparation of creatine from muscles, and if we then separate, by means of sulphuric acid, the baryta from the mother-liquid decanted from the creatine, and remove the volatile acids by evaporation of the fluid from which the sulphate of baryta has been separated by filtration, this sugar, together with sulphate of potash, crystallizes from the remaining acid fluid on the gradual addition of strong alcohol. The crystals of inosite may be readily picked out and separated from those of sulphate of potash after the mother-liquid has been removed by pressure, or the separation may be readily effected by boiling water, in which inosite is more soluble than sulphate of potash. Inosite may also be obtained, according to Scherer, without previous distillation, if we do not use quite sufficient sulphuric acid to throw down the whole of the baryta. On now shaking the solution with ether, till the liberated acids are separated, the inosite appears in beautiful crystals on the gradual addition of alcohol.

*Tests.*—Scherer<sup>2</sup> has given a very characteristic reaction for inosite, by which it may be distinguished from any other kind of sugar or carbohydrate. If we evaporate a solution of inosite, or a mixture containing that substance, almost to dryness on platinum foil, treat the residue with a solution of ammonia and a little chloride of calcium, and then carefully evaporate to dryness, a vivid rose-red tint is evolved, which allows us to recognize even 1-50th of a grain of inosite.

*Occurrence.*—This body has hitherto been only found in the flesh of the heart; Socoloff<sup>3</sup> sought in vain for it in the fluid from other muscular structures.



*Properties.*—This variety of starch presents itself as a glistening white granular matter, which, when freshly precipitated is translucent, and has a gelatinous and very swollen appearance: it is insoluble in water and in dilute acids; it is not converted into sugar either by dilute sulphuric acid or by diastase; and it is only by prolonged boiling with fuming hydrochloric acid that it yields a sweet fermentable substance. When heated to 200° it is converted into a gummy substance, which is soluble in water, but not in alcohol; at a higher temperature it fuses and

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 73, S. 322.

<sup>2</sup> Verhandl. d. phys.-med. Ges. zu Würzburg. Bd. 2, S. 112.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 81. S. 375.

burns with an odor resembling that of sugar. Paramylon is insoluble in ammonia, but dissolves in caustic potash, from which it may be again thrown down by acids. It is not colored blue by iodine.

*Composition.*—This body was discovered and analyzed by Gottlieb,<sup>1</sup> and was found to be isomeric with common starch :

Carbon,	.	.	.	.	.	12 atoms,	.	.	.	44.44
Hydrogen,	.	.	.	.	.	10 "	.	.	.	6.18
Oxygen,	.	.	.	.	.	10 "	.	.	.	49.38
										<hr/> 100.00

*Preparation.*—Paramylon was obtained by Gottlieb from the bodies of an infusorium, *Euglena viridis*. These animalcules after being freed as much as possible from other infusoria, were first treated with ether and spirit, and then with a boiling mixture of spirit and hydrochloric acid in order to remove the color; the residue was mixed with water and thrown upon a cotton filter which allowed the granules of paramylon to pass, but retained the investing membranes of the animals. The substance was purified by solution in potash-ley, and precipitation with hydrochloric acid.

#### CELLULOSE.— $C_{12}H_{10}O_{10}$ .

*Properties.*—In its purest state this substance forms a spongy mass, insoluble in all neutral menstrua, but very slightly soluble in alkaline solutions; it is convertible into dextrin both by sulphuric acid and by diastase.

If cellulose be treated with a mixture of four parts of concentrated sulphuric acid and one part of water, it swells on trituration into a clear jelly, which at first is stiff but gradually becomes quite fluid: on the addition of water or alcohol there is a deposition of white flakes which are insoluble in hot water, alcohol, and ether, but possess the remarkable property of being colored blue by iodine like starch; they differ, however, essentially from starch in this respect, that the iodine may be washed out with water and the blue color destroyed, which is not the case with starch. This product of the metamorphosis of cellulose has hence been named *amyloid*; its composition has been found to correspond with the formula,  $C_{48}H_{41}O_{41}$ . This substance is readily soluble in sulphuric acid, from which it may be again precipitated unchanged by water: it dissolves in strong nitric acid, without any development of gas; but on boiling there is a formation of oxalic acid: it only dissolves with difficulty in hydrochloric acid, from which it is not precipitated by ammonia; moreover, ammonia does not dissolve it. It swells in a strong solution of potash, and dissolves on the addition of water, from which it may be again thrown down by acetic acid. By the prolonged action of alkalis, cellulose is converted into a substance to which iodine communicates a dark violet, or almost black color. Rotten potatoes contain a ferment which dissolves and destroys the cellulose, but in no way affects the starch.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 81, S. 375.

The formation of this substance, and its reaction towards iodine, have been employed for the recognition of cellulose.

If, for instance, vegetable tissues, consisting of cellulose, be moistened with sulphuric acid and tincture of iodine, they assume a beautiful blue color, which, however, gradually disappears on the addition of water. Moreover, chloride of zinc converts cellulose first into a matter which is colored blue by iodine, then into sugar, and lastly into a humus-like substance.

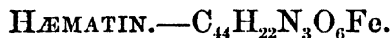
*Composition.*—According to Mulder, the composition of this substance is represented by the formula  $C_{24}H_{21}O_{21}$ ; but according to the more recent observations of Mitscherlich,<sup>1</sup> it is perfectly isomeric with starch.

*Occurrence.*—Allusion has already been made to its occurrence in certain of the lower animals. We can obtain cellulose in its purest form from the pith or young roots of the elder by treating them with various indifferent, as well as acid and alkaline solutions, in order to remove any adhering soluble matter. Swedish filtering paper is pure cellulose.

A carbo-hydrate has been found in some of the lower classes of animals whose composition and properties are very similar to those of the vegetable principle, *cellulose*. C. Schmidt<sup>2</sup> discovered it in the mantle of *Phallusia mammillaris*, one of the mollusca belonging to the Tunicata; and Löwig and Kölliker<sup>3</sup> have subsequently recognized it in the cartilaginous capsule of the simple Ascidiae, in the leathery mantle of the Cynthiæ, and in the outer tube of the Salpæ. The relation which this substance bears to chitin as well as to the animal organism generally, will be noticed in our remarks on chitin.

## COLORING MATTERS.

Unfortunately, even less is known of the chemical nature of the animal than of the vegetable pigments, so that we must still retain the irrational system of arranging them according to color.



### *Chemical Relations.*

*Properties.*—This substance is regarded as the peculiar red pigment of the blood-corpuscles; but unfortunately it is by no means certain whether it is a product of metamorphosis of the true coloring matter of the blood, or whether the substance prepared by us only bears the same sort of relation to that which exists in the blood-corpuscles as coagulated albumen bears to that principle in its fluid state. We cannot isolate it

<sup>1</sup> Ber. d. Akad. d. Wiss. zu Berlin. 1850, S. 102-111.

<sup>2</sup> Zu vergl. Physiol. der wirbellosen Thiere. 1845. S. 62 [or Taylor's Scientific Memoirs, vol. 5, p. 34.—G. E. D.]

<sup>3</sup> Ann. de Scienc. Nat. 3 Sér. T. 5, pp. 193-232.

in its soluble state from the globulin of the blood-corpuscles; hence we are only acquainted with it in its coagulated (and essentially modified) condition. In a state of purity it occurs as a dark-brown, slightly lustrous mass, which, on trituration, adheres to the pestle; it is devoid of taste and smell, and is insoluble in water, alcohol, ether, acetate of oxide of ethyl, and fatty and volatile oils; Mulder, however, regards it as slightly soluble in fatty and ethereal oils.

Hæmatin dissolves very readily in weak *alcohol* to which sulphuric or hydrochloric acid has been added, forming a brown solution, which, on saturation with an alkali, assumes a blood-red color. *Water*, acidulated with the same acids, exerts no solvent power on hæmatin, and consequently a precipitation is induced by the addition of water to alcoholic solutions of this substance. Concentrated sulphuric and hydrochloric acids do not dissolve hæmatin, but they abstract a little of the iron. After trituration with sulphate of soda, it dissolves for the most part in water. Even very dilute solutions of the caustic *alkalies* or their carbonates in water or alcohol dissolve hæmatin in almost every proportion. A potash-solution, boiled and then saturated with an acid, yields a form of hæmatin which is no longer soluble in a mixture of alcohol and ammonia. The potash-solution, on boiling, assumes first a dark red, and subsequently a green tint. The ammoniacal solution gives off its ammonia during evaporation; moreover, hæmatin does not absorb ammoniacal gas. The color of the ammonia-solution of hæmatin is not affected by *carbonic acid*, *oxygen*, or *nitric oxide*; sulphurous acid gives it a bright red tint, and sulphuretted hydrogen makes it slightly darker.

Hæmatin is completely precipitated from its ammonia-solution by the *salts of oxide of silver, of lead, and of copper*; if the solution of hæmatin in alcohol, acidulated with sulphuric acid, be boiled with oxide of lead, it becomes entirely decolorized.

When *heated* in an enclosed space, hæmatin puffs up, and, without melting, yields empyreumatic ammoniacal vapors and a reddish-brown oil, and leaves a rather small porous charcoal, which on combustion yields a red ash. *Phosphorus* and *sulphate of protoxide of iron* may be boiled with hæmatin without in any way affecting it.

Treated with concentrated *nitric acid* in the cold, it dissolves into a brown fluid, and develops nitrous acid; when boiled with this acid it is entirely destroyed.

If *chlorine* be allowed to act on hæmatin mixed with water, all the iron of the hæmatin dissolves as perchloride of iron, and there is a deposition of white flocculi, which are soluble in alcohol and ether but not in water, develop a little chlorous acid when dried (at 100°), and then form a light straw-colored powder. This powder is unaffected by hydrochloric acid, but dissolves in alkalies, forming a reddish solution; according to Mulder, it consists of chlorous acid and hæmatin freed from its iron. If chlorine gas be passed over dry hæmatin, they unite and form a dark-green compound which is soluble in alcohol, exerts no action on vegetable colors, is unaffected by acids and alkalies, but which, when warmed with hydrosulphate of ammonia, assumes a red color.

On passing dry *hydrochloric acid gas* over dry hæmatin, there is formed a violet mass, which is soluble both in water and alcohol, communicating to those fluids a red color and an acid reaction.

If hæmatin be allowed to remain for some time in contact with pure concentrated sulphuric acid, and the fluid be then diluted with water, there is a development of hydrogen gas, and sulphate of protoxide of iron is taken up in solution. By a repetition of this process the whole of the iron, with the exception of a mere trace, may be removed from the hæmatin, without depriving it of its properties and without altering its elementary composition, as far as the relative amounts of the carbon, hydrogen, nitrogen, and oxygen are concerned.

We are indebted to Mulder and van Goudoever<sup>1</sup> for the preparation of hæmatin free from or poor in iron; Sanson and Scherer<sup>2</sup> had, however, previously observed that concentrated sulphuric acid could extract all the iron from the clot or the residue of the blood-corpuscles, without affecting its dark-brown color.

*Composition.*—Mulder<sup>3</sup> has calculated, from his analyses, the formula we have given for hæmatin, according to which it contains:

Carbon, . . . . .	44 atoms,	. . . . .	65.347
Hydrogen, . . . . .	22 “ . . . . .	. . . . .	5.445
Nitrogen, . . . . .	3 “ . . . . .	. . . . .	10.396
Oxygen, . . . . .	6 “ . . . . .	. . . . .	11.881
Iron, . . . . .	1 “ . . . . .	. . . . .	6.931
			100.000

Mulder's analyses of hæmatin free from iron coincide with the formula  $C_{44}H_{22}N_3O_6$ . From the chloride of hæmatin Mulder calculates that the atomic weight of hæmatin is 5175.

*Chloride of hæmatin*, formed from dry chlorine gas and hæmatin, consists of 1 equivalent of hæmatin, and 6 equivalents of chlorine; how this combination may be supposed to be formed, is a point on which at present we can offer no conjecture. The compound obtained from dry hydrochloric acid gas and hæmatin consists, according to Mulder, of 2 equivalents of hæmatin and 3 equivalents of hydrochloric acid; on exposing this substance to a heat of  $100^\circ$ , it loses half its acid, and then consists of 4 atoms of hæmatin and 3 atoms of acid. In the combinations of hæmatin with metals it appears from an experiment of Mulder's that 1 atom of hæmatin is combined with 1 atom of base.

The question—in what condition does the iron exist in the blood, or on what iron-compound is its red color dependent? is one that has long engaged the attention of chemists and physiologists. Without considering that, with an equal right we might inquire into the causes of the color of indigo, carmine, or peroxide of iron, it was universally believed that the blood's color must depend on the last-named substance, and consequently, all experiments on the subject were instituted with the view of ascertaining in what state of combination the peroxide of iron lay concealed. It would be superfluous for us to notice the different views regarding the combinations in which the peroxide of iron has been supposed to exist in the blood. We must not, however, omit all notice of the circumstance, that a discovery of Engelhardt's showed the fallacy of these views, for he ascertained that the iron of the blood might be pre-

<sup>1</sup> Journ. f. pr. Ch. Bd. 325, S. 186, ff.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 30.

<sup>3</sup> Journ. f. pr. Ch. Bd. 28, S. 340.

precipitated by alkalis and liver of sulphur, if chlorine gas had been previously, and for some time, passed through the blood; and this led him to the somewhat illogical conclusion that the iron could not be oxidized, but must exist in a metallic state in the blood; for Rose's discovery that the precipitation of peroxide of iron and other metallic oxides may be prevented by all the non-volatile organic acids, shows that notwithstanding Engelhardt's experiment, the iron may be contained in the blood in the state of peroxide. Finally, Lecanu discovered the true coloring matter of the blood, the hæmatin; and as almost all the iron of the blood is contained in this substance, attempts were again made to refer the color of this pigment to peroxide of iron. But we know, from the experiments of Scherer, Sanson, and Mulder, that the iron must be contained in some other combination than in direct combination with oxygen, and that the iron may be abstracted from the red blood-pigment without affecting its color. That the iron is directly combined with the group of atoms constituting hæmatin, is not a probable view; at present, however, we are in possession of no facts throwing any additional light on the nature of the iron-compound.

The white body formed by the action of chlorine and water on hæmatin, was found by Mulder to be devoid of iron, and to be composed in accordance with the formula,  $C_{44}H_{22}N_3O_6 + 6ClO_3$ .

*Preparation.*—We treat blood with about eight times its volume of a solution of sulphate of soda or chloride of sodium, filter it, and wash the residue on the filter as thoroughly as possible with the same saline solution; the residue thus almost completely freed from serum, or, in other words, the mass of the blood-corpuscles, is dissolved in water, and coagulated by the application of heat; the washed, dried, and finely triturated coagulum is now boiled with spirit containing sulphuric acid, till the fluid passes through a filter in a decolorized state. This filtered fluid, which in the mass presents a brownish-red tint, after being saturated with ammonia, deposits sulphate of ammonia and a little globulin; these being removed by filtration, the fluid is evaporated to dryness; the solid residue is extracted with water, alcohol, and ether, and in order to effect the complete removal of any adhering globulin, is again dissolved in spirit containing ammonia; the solution is then filtered, evaporated, and the residue extracted with water.

*Tests.*—If from any suspicion of the presence of blood we wish to examine a fluid for hæmatin, it is by far the best plan to employ the microscope, and by its means to endeavor to detect blood-corpuscles, or their fragments. It only rarely happens, in certain exudations or saturated masses in which blood-corpuscles are no longer present, that we can with certainty recognize the red pigment of the blood, since its quantity is so small, that we can scarcely obtain enough, by the methods we have given, to apply any tests to it.

That the *hæmatoidin* discovered, or at least first accurately investigated by Virchow<sup>1</sup> (the same substance which has also been named *xanthose*), is not perfectly identical with hæmatin is obvious from Virchow's experiments; but the occurrence of this substance in sanguinous extravasations, whose metamorphoses have been most admirably traced out

<sup>1</sup> Arch. f. pathol. Anat. u. s. w. Bd. 1, S. 383-445.



by Zwicky, Bruch, and Virchow, denotes as decidedly as chemical experiments could do, that it is formed from hæmatin; moreover, several of its properties indicate its close affinity with the last-named substance.

*Hæmatoidin* occurs in an amorphous condition in granules, globules, and jagged masses, as well as in perfectly formed crystals of the monoclinometric system; these latter are oblique rhombic prisms, not unlike crystals of gypsum, but frequently are almost perfect rhombohedra; they are strongly refractive and transparent, are of a yellowish-red, red, or ruby color, and are insoluble in water, alcohol, ether, acetic acid, dilute mineral acids, and alkalies. I have sometimes seen the smaller and less deeply colored crystals dissolve in alcohol containing sulphuric acid or ammonia, and be again precipitated by neutralization of the fluid; this is, however, not invariably the case. Virchow has very carefully examined the behavior of this body with concentrated alkalies and mineral acids; these agents, however, do not act in precisely the same manner on all specimens of this pigment; on the addition of hydrate of potash, a fiery red tint is developed, the mass becomes gradually loosened in its texture, and becomes disintegrated into red granules which at length dissolve; on neutralizing the alkali the substance is, however, not again precipitated. When a concentrated mineral acid, sulphuric acid, for example, acts on it, it causes the sharp outlines of the crystals to disappear; and the color of the roundish fragments, after first becoming brownish-red, passes through successive shades of green, blue, and rose-tint, till it finally terminates in a dirty yellow. Iron may sometimes, but not always, be detected in the acid fluid containing the decomposed hæmatoidin.

Hæmatoidin may always be found in the sanguineous extravasations occurring in consequence of the bursting of the Graafian vesicles at the periods of menstruation or conception, and frequently occurs in old extravasations in the brain, in obliterated veins, hæmorrhagic infarctus of the spleen, in subcutaneous sugillations, and in purulent abscess of the extremities (Virchow). It appears from Virchow's observations that these crystals may form from seventeen to twenty days after the occurrence of the extravasation. Kölliker<sup>1</sup> has observed the formation of crystals of this nature within the corpuscles in the blood of certain fishes; these crystals were, however, soluble in acetic acid, potash, and nitric acid.

Although every care and precaution have been taken, both Virchow and I have failed in obtaining these crystals of modified hæmatin either from solutions of blood or of hæmatin itself; but yet those who still assign an important part in the animal body to vital forces, must grant that under the necessary conditions, hæmatoidin may be produced out of the body from hæmatin, since this kind of metamorphosis occurring in extravasations in all respects exhibits the character of a disintegration, that is to say, of a purely physical and chemical process. Moreover, Kölliker's observation gives us room to hope that we may be able to obtain crystallizable hæmatin or hæmatoidin from the blood of the lower animals—fishes, for example,—so as to submit it to an accurate chemical examination.

<sup>1</sup> Zeitschr. f. wiss. Zoologie, Bd. 1, S. 266.

*Physiological Relations.*

*Occurrence.*—Hæmatin has hitherto only been found in the blood-corpuscles of the higher animals. Intimately united with globulin, it forms the viscid, fluid contents of the colored blood-cells.

Berzelius found 0.38% of metallic iron in the dried blood-corpuscles of man or the ox; now as Mulder has found 6.64% of iron in hæmatin, a simple calculation shows that in the blood-corpuscles there are contained 5.72% of hæmatin, independently of fat, globulin, salts, and biliary matter: hence, in fresh blood in which the red blood-corpuscles on an average = 12.8%, there would be contained 0.732% of hæmatin. If we calculate from Becquerel's results, according to which 1000 parts of blood contain 0.565 parts of iron and 141.1 of blood-corpuscles, we obtain a very similar result, namely, that 100 parts of blood-corpuscles contain 6.02 of hæmatin. It is obvious that such calculations can only lead to approximate results; attempts have certainly been made to effect a direct determination of the amount of hæmatin in the blood; but the method of separating it is as yet too uncertain to admit of our placing much reliance on the numbers which have been obtained. The amount found in the blood by Lecanu, namely 0.227%, was obviously too small, while Simon's number, 0.718%, approximates closely to the calculated quantity.

By treating defibrinated calves' blood with chloride of sodium, Schmidt obtained the corpuscles in a state of purity; and after incineration, found in them 1.179% of peroxide of iron, hence (according to Mulder's analysis of hæmatin) they would contain 12.41% of this ingredient; in repeating Schmidt's experiment with ox-blood I obtained 9.076 and 10.94% of peroxide of iron—results which corresponded tolerably well with that which he found. The great difference which presents itself between these results of direct experiment, and the results of pre-indicated calculations, admits of an easy explanation; in the latter case, the blood-corpuscles are calculated more or less in accordance with their true constitution in the blood, while in our experiments, the process by which we purify the blood-corpuscles—their treatment with a solution of chloride of sodium or sulphate of soda—abstracts from them a portion of their globulin, and all the soluble salts; when treated with saline solutions, the corpuscles lose, in accordance with the laws of endosmosis, not only water, but also a part of their soluble globulin; while the treatment of the coagulated corpuscles with water, alcohol, and ether, abstracts from them all soluble salts, and the fat, which in itself amounts, according to my investigations, to at least 2%.

The ratio of the hæmatin to the blood varies in *diseases* for the most part with the number of the blood-corpuscles; but whether the ratio of the hæmatin to the globulin of the blood-corpuscles be constant, or whether the hæmatin be liable to greater variations than the globulin, are questions which in the present state of organic analysis it is impossible to answer.

*Origin.*—There is nothing in the chemical constitution of hæmatin which throws any light on the mode of its formation; we do not know whether it is directly formed from the constituents of the food or from

the products of metamorphosis of effete tissue; and we have no certain knowledge regarding the part of the organism in which it is produced. The chyle certainly contains iron, and hæmatin exists in the thoracic duct; but iron is not hæmatin, and the small quantity of the last-named substance may have passed from the blood through the mesenteric glands into the chyle, or may have arisen from the blood-corpuscles which have passed with the splenic lymph into the chyle. If the formation of hæmatin took place in the chyle it would not be after prolonged fasting that we should find it richest in this substance. Chemistry, as we have already observed, affords us no assistance in reference to the formation of this body; we must, therefore, at present, confine our attention to physiological facts, in order that we may obtain a safe starting-point for further chemical inquiries.

Most physiologists of the present day coincide in the opinion that the red blood-corpuscles are developed from the colorless ones; but whether they regard the former as nuclei of the latter, or as independent cells produced from them—whether they adopt the views of H. Müller,<sup>1</sup> of Gerlach,<sup>2</sup> or of Kölliker<sup>3</sup>—they must in any case admit that the red pigment of the blood is primarily formed within the enveloping membrane of the cell. Further, physiological inquiry demonstrates, almost beyond a doubt, that the blood-pigment is first formed in the perfected cells, and, moreover, affords us some indication, however indistinct, of the source from whence this pigment may possibly have been produced. Nasse, Hünfeld, and others, have proved that the granular matter visible in many of the colored blood-corpuscles is merely fat; indeed in the yolk-cells, in the young blood-corpuscles of the amphibia [in their embryonic state] we find not only roundish but also angular granules soluble in ether, which can hardly be anything else than stearin. Henle and H. Müller refer the primary origin of the colorless blood-corpuscles to the fat which is recognizable as a fine granular (almost cloudy) matter in the minutest lacteals. We have already mentioned that the fat stands in a certain relation to the functions of the liver; the beautiful investigations of E. H. Weber and Kölliker have, however, now demonstrated that large quantities of blood-corpuscles are always formed in the liver in the foetal state, and during the hibernation of certain animals, and therefore at periods when this organ secretes little or no bile, but when fat is accumulated in it.

Moreover, an unprejudiced examination of the development of the chick within the egg leads to the assumption that the fat takes a part in the formation of hæmatin; and if physiological facts can be adduced in favor of this hypothesis, there are at all events no chemical objections to it. As it is obvious that the coloring matter can only be formed when there is free access of oxygen, namely in the vessels, and as the oxygen doubtless contributes materially to its production, we cannot suppose that it is formed from protein, which is a substance rich in oxygen, or from sugar; hence there is hardly any other substance than the fat from which a process of oxidation could yield hæmatin.

Our present assumption of the formation of hæmatin from fat is to be

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 3, S. 204-278.

<sup>2</sup> Ibid. Bd. 7, S. 70-90.

<sup>3</sup> Ibid. Bd. 4, S. 112-160.

regarded merely as an hypothesis based on one or two physiological facts, which may possibly admit of a very different interpretation; it is only intended to serve as a means of directing our attention in a definite direction in the investigation of this subject.

*Uses.*—The constant occurrence of hæmatin in the blood-corpuscles indicates that this body takes an important part in the metamorphosis of the animal tissues. All sorts of conjectures have been hazarded regarding its function in the blood, and it has been especially supposed to be connected with the process of respiration. In point of fact, however, it is unnecessary to consider any hypothesis, until it has been satisfactorily ascertained whether the hæmatin in question actually stands in the same relation to the true pigment of the blood as coagulated to non-coagulated albumen, or whether artificially prepared hæmatin is altogether a product of decomposition of the actual pigment.\* If hæmatin has the same composition as that which we prepare artificially, and if the only difference be that it exists in a soluble form in the blood-corpuscles, there is at once an end to all those very imaginative hypotheses which assume that the iron takes a great share in the process of respiration, and that it is the conveyer of oxygen to the blood.

The experiments of Bruch<sup>1</sup> on the action of gases on the color of the blood, and the observations of Harless,<sup>2</sup> regarding the gradual destruction of the corpuscles of frogs' blood, certainly indicate that there is a chemical action between the blood-corpuscles and their contents on the one hand, and the inspired oxygen on the other, in which action the hæmatin doubtless participates.

The observations of Hannover,<sup>3</sup> which show that persons whose blood is very deficient in red corpuscles (chlorotic persons) exhale as much carbonic acid as healthy persons, seem on the other hand to contra-indicate a direct relation between the blood-corpuscles or blood-pigment, and oxidation in the blood. We must, therefore, give up for the present all attempts at understanding the function of the blood-pigment.

The question as to what becomes of the hæmatin when the blood-corpuscles and their contents undergo disintegration, is one which for a long time was enshrouded in perfect obscurity, but on which some light has now been thrown by Virchow's admirable investigations on hæmatoidin. The occurrence, in a crystalline form, of this substance, which is undoubtedly derived from the blood-pigment, and its different behavior towards the same reagents, indicate that, notwithstanding its crystalline arrangement, it continues to undergo changes which give rise to a substance perfectly similar to, if not identical with, bile-pigment or melanin. Although the subject is still far from being satisfactorily settled, Virchow was the first who by his pathologico-histological and chemical investigations prominently brought forward definite facts which have afforded the first solid groundwork for the hypothesis which was long since propounded, that hæmatin might be transformed into cholepyrrhin.

In reference to this point we would specially direct attention to Vir-

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 3, S. 308.

<sup>2</sup> Ueber den Einfluss der Gase auf die Blutkörperchen von Rana tempor. Erlangen, 1846.

<sup>3</sup> De quantitate acidi carbonici ab homine sano et aegroto exhalati. Hauniæ, 1845.

chow's ingenious treatise, in which he endeavors to strengthen the view regarding this metamorphosis by means of a simple induction based on direct observation. It has unfortunately hitherto been found impossible to separate hæmatoidin in so pure a state and in sufficient quantities as to admit of its being subjected to a rigid chemical investigation. From Virchow's investigations it is, however, apparent that the physician must also lend his help for the advancement of pathological and physiological chemistry; for without the aid of pathological histology,—without a judicious application of the microscope,—the chemist could not have succeeded in discovering hæmatoidin any more than in detecting oxalate of lime in normal urine; without such aid the chemist could never have conceived an idea of the metamorphosis of the pigments in the animal body. As long as the physician contents himself with borrowing mere hypotheses from chemists, without being himself practically familiar with chemical science, he can never hope to gain the advantages which it is capable of affording; in this respect he resembles the agriculturist, who can never expect to raise his pursuit to the dignity of a science until he has learned the practical application of the principles of chemistry.

## MELANIN.

### *Chemical Relations.*

*Properties.*—Melanin forms either a black, cohesive mass, or a blackish-brown powder; it is devoid of smell and taste; when stirred in water it continues to float for some time, but is insoluble both in water and in alcohol, in ether, in dilute mineral acids, and in concentrated acetic acid; it dissolves, after prolonged digestion, in a dilute solution of potash, from which it is again precipitated with a light-brown color by hydrochloric acid; it is decomposed when boiled with concentrated nitric acid, but it is not affected even by the very prolonged action of chlorine. It is a conductor of electricity, is incapable of fusing, may be ignited in the air, and burns with a vivid light, the charcoal continuing to smoulder till it is reduced to a whitish-yellow ash consisting of chloride of sodium, lime, bone-earth, and a little peroxide of iron. By dry distillation it yields an empyreumatic substance, and carbonate of ammonia. According to Gmelin this pigment is rendered paler, and is partially dissolved by chlorine-water, the undissolved portion becoming again of a dark-brown color on the addition of potash.

Whether the black crystals which have been found by Mackenzie,<sup>1</sup> Guillot,<sup>2</sup> and Virchow,<sup>3</sup> in melanotic masses are or are not identical with melanin, is a question which, with our present very imperfect knowledge of this pigment, must still remain undecided. Virchow found these crystals to be flat rhombic tablets with extremely acute angles.

*Composition.*—Scherer<sup>4</sup> gives the following as the mean result of three analyses of this body:

<sup>1</sup> A Practical Treatise on Diseases of the Eye. Lond. 1835, p. 663.

<sup>2</sup> Arch. gén. de Méd. 4 Sér. T. 7, p. 166.

<sup>3</sup> Arch. f. Pathol. Anat. u. s. w. Bd. 1, S. 399.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 6.

Carbon,	.	.	.	.	.	.	.	.	.	58.084
Hydrogen,	.	.	.	.	.	.	.	.	.	5.917
Nitrogen,	.	.	.	.	.	.	.	.	.	13.768
Oxygen,	.	.	.	.	.	.	.	.	.	22.231
										<hr/> 100.000

As we are neither acquainted with the atomic weight of this body, nor with any of the products of its decomposition, we cannot attempt to construct a hypothetical formula for it. In the pigment from the choroid coat of the eye I found 0.254<sup>o</sup> of iron.

The black pigment which is often deposited as a morbid product in the lungs presents great differences of composition. In two different cases which C. Schmidt<sup>1</sup> analyzed he found :

Carbon,	.	.	.	.	72.95	.	.	.	.	66.77
Hydrogen,	.	.	.	.	4.75	.	.	.	.	7.33
Nitrogen,	.	.	.	.	3.89	.	.	.	.	8.29
Oxygen,	.	.	.	.	18.41	.	.	.	.	17.61
					<hr/> 100.00					<hr/> 100.00

*Preparation.*—The best method of obtaining this body is from the eye, by removing the retina, and detaching the choroid coat from the sclerotic. The choroid coat must be placed in a clean rag, and the coloring matter washed out with pure water, just as the starch-granules in the preparation of gluten are washed out through linen bags; the pigment remains for a long time suspended in the water, from which, however, it may be readily removed by filtration, or the fluid may be evaporated and the residue extracted with water.

*Tests.*—The physical properties of this body are so characteristic, that it is easy to recognize and to separate it; generally, however, it only occurs in such small quantities that it is impossible to distinguish whether the object in question is identical with the melanin of the eye, especially as we still know comparatively little regarding the chemical characters of this last-named substance. No conclusions regarding the presence of black pigment can be drawn from mere color and insolubility in different menstrua, since, as Jul. Vogel<sup>2</sup> was the first to observe, the tissues may be infiltrated with sulphide of iron, from which, however, the black pigment may very readily be distinguished by means of acids.

#### *Physiological Relations.*

*Occurrence.*—This pigment exists as a thick investment on the choroid coat of the eye. Whether it also occurs in other parts of the animal organism, is a point which cannot be decided, since the other pigments of the same color in morbid depositions either have not been accurately analyzed, or from their very small quantity do not admit of analysis; as for instance, the pigment of the black bronchial glands, of the *rete mucosum seu malpighianum* of the negro, of melanotic tumors, of the black serum which has been occasionally observed, and of pulmonary tissue in certain cases.

In the choroid coat the melanin is enclosed in peculiar hexagonal

<sup>1</sup> Vogel's Pathol. Anat. S. 161 [or English Translation, p. 192].

<sup>2</sup> Pathol. Anat. S. 163 u. 311 [or English Translation, pp. 194 and 396].

cells, but in the coats of the bloodvessels of frogs and other amphibia it is found in jagged ramifying cells. In other parts of the animal body—in melanotic tumors for instance—it occurs, however, merely scattered among other cells or tissues. Whether granular cells, when becoming obsolete (such for example as we find in old exudations), contain actual melanin, is a question which must still remain undecided. Sanguineous extravasations are, however, not unfrequently converted into a mass, which is colored perfectly black by black pigment.

*Origin.*—The large quantity of iron contained in this pigment indicates that it takes its origin from the hæmatin. We cannot recognize such a conversion by chemical means, till we are able to demonstrate that pathological depositions of pigment contain true melanin. Whatever view we may adopt regarding the production of the black-colored inflammatory globules, we must at all events agree with Bruch<sup>1</sup> that they contain blood-pigment and the rudiments of blood-corpuscles, even if we do not, like Hasse,<sup>2</sup> H. Müller, and Pestalozzi,<sup>3</sup> see true blood-corpuscles in these cells; if we examine the expectoration in a case of pneumonia in which resolution is very gradually progressing, we find, on making a perfectly unprejudiced observation, very many of these cells which have the exact color of blood-corpuscles. Virchow<sup>4</sup> has very accurately traced, by microscopical examination, the conversion of isolated coagula in obliterated veins into amorphous and crystalline pigment, and from these morphological investigations it can hardly be doubted, that at all events the melanin of morbid products is formed from the hæmatin. Kölliker<sup>5</sup> has moreover convinced himself that in the blood-corpuscles enclosed in the enveloping membrane, the hæmatin affords the matter from which the black pigment in the granular cells is formed. Hence it only remains for the chemist to continue his investigations on this subject, in order to obtain perfectly satisfactory scientific proof of this metamorphosis.

*Uses.*—That the use of pigment in the choroid coat is principally to render the eye achromatic, is sufficiently obvious from the principles of physics. We are ignorant of the uses which it serves in the walls of the bloodvessels in the amphibia.

## BILE-PIGMENT.

### *Chemical Relations.*

*Properties.*—This substance, like so many of the pigments, belongs to that vast group of bodies, whose chemical properties have never been thoroughly investigated; this is partly dependent on the circumstance that we can only procure it in very small quantity, and partly on its extreme instability, for not only does it occur in the animal organism

<sup>1</sup> Untersuch. zur Kenntniss des körnigen Pigments der Wirbelthiere. Zurich, 1844. S. 42 ff, and Zeitschr. f. rat. Med. Bd. 4, S. 24 ff.

<sup>2</sup> Zeitschr. f. rat. Med, Bd. 4, S. 1-15.

<sup>3</sup> Ueber Aneurismata spuria der kleinen Hirnarterien u. s. w. Würzb. 1849.

<sup>4</sup> Arch. f. Pathol. Anat. u. s. w. Bd. 1, S. 401.

<sup>5</sup> Zeitschr. f. wiss. Zoologie. Bd. 1, S. 260-267.

under various modifications, but it is at once changed by the simplest chemical treatment. The most frequent modification which the primary substance of the bile-pigment in the higher animals appears to present, is the *brown pigment*, the *cholepyrrhin* of Berzelius, and the *biliphæin* of Simon. It occurs as a reddish-brown, non-crystalline powder, devoid of taste and smell; it is insoluble in water, very slightly soluble in ether, and more so in alcohol, to which it communicates a distinct yellow tint; it is more soluble in caustic potash than in caustic ammonia, the alkaline solutions being at first of a clear yellow color, but on exposure to the air gradually changing to a greenish-brown tint. It is on this modification of the bile-pigment that the well-known changes of color which occur in some of the animal fluids are dependent. The yellow solution of this pigment when gradually treated with *nitric acid* (and especially, according to Heintz,<sup>1</sup> when this reagent contains a little nitrous acid), first becomes green, then blue (which, however, can hardly be detected in consequence of its rapid transition into violet), and red; after a considerable period the red again passes into a yellow color; by this time, however, the bile-pigment is entirely changed. On the addition of *hydrochloric acid* to a potash-solution, the pigment is precipitated with a green tint; this precipitate forms a red solution with nitric acid, and a green solution with the alkalies, and appears to be perfectly identical with the green modification of bile-pigment. The coloring-matter contained in fresh bile is colored green by acids; as Gmelin found that this coloration did not take place without the free access of oxygen, it is highly probable that most of these changes of color are dependent on a gradual oxidation. Chlorine gas acts on this pigment in the same manner as nitric acid, but rather more rapidly; large quantities of chlorine completely bleach the pigment, and precipitate it in a white flocculent deposit.

This brown pigment has a strong tendency to combine with bases,—not merely with alkalies, but also with metallic oxides and alkaline earths. It forms insoluble compounds with the alkaline earths—a circumstance which has often led to the idea that this substance is insoluble.

The *green pigment*, the *biliverdin* of Berzelius, is a dark-green amorphous substance, devoid of taste and smell, insoluble in water, slightly soluble in alcohol, but dissolving in ether with a red color; it dissolves in fats, hydrochloric acid, and sulphuric acid with a green color, and in acetic acid and the alkalies with a yellowish-red tint. On exposure to heat, this body undergoes decomposition without fusing, and without giving off any appreciable quantity of ammonia, leaving a little charcoal. Berzelius regards this substance as perfectly identical with the chlorophyll of leaves, and believes that he has found all three modifications of this substance in different specimens of bile. This green pigment no longer undergoes changes of color on the addition of nitric acid, although we occasionally meet with green bile-pigment still possessing this property. On treating bile-pigment with alkalies or acids, its properties are usually at once changed, partly on account of its entering into various combinations with these substances, and partly from the extreme facility with which it becomes decomposed.

Hence it is that the statements regarding the properties of this sub-

<sup>1</sup> Müller's Arch. 1846, S. 399-405.



stance present such striking differences, as may be seen by a comparison of the writings of Berzelius,<sup>1</sup> Scherer,<sup>2</sup> Hein,<sup>3</sup> Platner,<sup>4</sup> and others.

Berzelius also found in the bile a substance occurring in small reddish-yellow crystals, soluble in alcohol, to which he has given the name of *bili-fulvin*. I have obtained it in solution, but have never succeeded in isolating it in the solid state; singularly enough, I have often found it in the bile precipitated with neutral and basic acetate of lead; hence it appears either not to be precipitated by these metallic salts, or (which is more probable) to redissolve in an excess of the basic salt.

The *bili-fulvin* of Virchow must not be confounded with the *bili-fulvin* of Berzelius; the former seems to be identical with the *hæmatoidin* also discovered by Virchow. Virchow<sup>5</sup> found *hæmatoidin* constantly present in the extravasated blood consequent on the bursting of a Graafian vesicle in menstruation or conception, and he often noticed it in old extravasations of blood in the brain, in obliterated veins, in hæmorrhagic infarctus of the spleen, in subcutaneous sugillations, and in abscesses in the extremities. It appears from Virchow's investigations, that these crystals are formed in from 17 to 20 days after the extravasation has occurred.

*Hæmatoidin* occurs in an amorphous condition in granules, globules, and jagged masses, as well as in perfect crystals belonging to the monoclinic system. These crystals are oblique rhombic prisms, not unlike crystals of gypsum: they often, however, occur as nearly perfect rhombohedra; they are strongly refractive and transparent, of a yellowish-red, red, or ruby-red. color; they are insoluble in water, alcohol, ether, acetic acid, dilute mineral acids and alkalies. I have on several occasions seen the smaller and lighter colored crystals dissolve in alcohol containing sulphuric acid or ammonia, and again precipitated by ammonia; this, however, is not always the case. Virchow has accurately studied the behavior of this body with concentrated alkalies and mineral acids: these reagents, however, do not seem to act uniformly on all specimens of *hæmatoidin*; on the addition of hydrated potash the pigment usually assumes a glowing red tint, the mass gradually separating and breaking up into red granules, which slowly dissolve; the substance is, however, not again precipitated by the neutralization of the alkali. If we allow concentrated mineral acids (sulphuric acid for instance) to act on *hæmatoidin*, the clear outlines of the crystals disappear and the color of the roundish fragments passes first into a brownish-red, then into a green, a blue, and a purple tint, and finally merges into a muddy yellow. Iron may occasionally, but by no means invariably, be found in the acid fluid that is formed during the decomposition of *hæmatoidin*.

Virchow<sup>6</sup> subsequently discovered peculiar reddish-yellow, elongated crystals, which were either acicular or arranged in zigzag rows or bars in the bile of persons who had suffered from cancer of the liver or retention of the bile consequent on catarrh of the gall-bladder; these crystals ranged from 0.005 to 0.010''' in length, while the breadth scarcely admitted of measurement. They dissolve readily in caustic

<sup>1</sup> Lehrb. d. Ch. Bd. 9, S. 281-286.

<sup>2</sup> Journ. f. pr. Ch. Bd. 40, S. 47-56.

<sup>3</sup> Arch. f. path. Anat. Bd. 1, S. 383-445.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 377.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 51, S. 115.

<sup>6</sup> Op. cit. pp. 427-431.

potash, but are not again precipitated by the addition of acids. Acetic acid has no effect upon the crystals; concentrated sulphuric acid makes them assume a somewhat darker color, and gradually destroys them; moderately dilute nitric acid exerts little action on them. Besides these zigzag crystals, to which (as has been already mentioned) Virchow assigned the name of "bilifulvin," he sometimes also found crystals which were perfectly similar to those of hæmatoidin both in form and color. While Virchow has repeatedly pointed out the great similarity which exists between this bilifulvin and hæmatoidin, Dr. Zenker<sup>1</sup> (of Dresden) has recently discovered that if these substances are not identical, there is at all events the closest relationship between them, since he has proved that the bilifulvin may be very readily converted into hæmatoidin. For if we allow bile containing bilifulvin to stand for a long time (several weeks) in contact with ether, the zigzag crystals of bilifulvin disappear, and in their place we have crystals of hæmatoidin (some of which are of very considerable size), which in their form, color, and *micro-chemical reactions* are precisely similar to the crystals of hæmatoidin formed within the body. Funke<sup>2</sup> has arrived at the same result simultaneously with, but independently of, Zenker. He allowed some bile containing bilifulvin to dry; on again moistening it, he found that the zigzag crystals were replaced by light-red crystals of hæmatoidin. By a series of careful investigations Zenker has arrived at the conclusion that as hæmatoidin is always formed when blood in a stagnating state occurs in the body, so this substance, bilifulvin, is produced wherever bile stagnates.

*Composition.*—With our present ignorance of bile-pigment in its pure unchanged state, it is not to be wondered at that its elementary composition is still unknown. Bile-pigment has been analyzed both by Scherer and Hein, but it is obvious from their analyses that they have examined very different substances, and Scherer has especially shown that the pigment which he examined loses much carbon and hydrogen by the action of air, alkalies, and acids. From 7 to 9% of nitrogen has been found in bile-pigment.

*Preparation.*—Till recently the ordinary mode of preparing bile-pigment consisted in the extraction, by water and ether, of biliary calculi, consisting for the most part of this constituent; the residue thus obtained does not, however, generally possess the power of dissolving in alcohol, for (as Bramson<sup>3</sup> has very correctly shown, and as any unprejudiced observer may easily convince himself) it exists in a state of insoluble combination with lime, even in those concretions which for the most part consist of cholesterin.

The mode of investigation which Bramson adopted, and which I have often repeated, appears to me to leave no doubt regarding the correctness of his views, which moreover receive further confirmation from the analyses of biliary concretions made by Schmid<sup>4</sup> and Wackenroder.<sup>5</sup>

Berzelius prepares biliverdin from ox-gall by precipitating the alcoholic

<sup>1</sup> In a private communication. The details are to be published in Henle's Zeitsch. f. rat. Med.

<sup>2</sup> In a private communication.

<sup>3</sup> Zeitschr. f. rat. Med. Bd. 4, S. 193-208.

<sup>4</sup> Arch. der Pharm. Bd. 41, S. 291-293. <sup>5</sup> Ibid. S. 294-296.

extract with chloride of barium; the precipitate is first washed with alcohol, and afterwards with water, and then decomposed with hydrochloric acid, which extracts the baryta; the fat is removed by ether from the residue, which is then dissolved in alcohol.

Platner precipitates the bile-pigment by digesting the bile with hydrated protoxide of tin; the light-green deposit which is formed, after being well washed with water, is shaken with spirit containing sulphuric acid, and filtered; the pigment is thrown down in the form of a green flocculent precipitate on the addition of water to the filtered green solution.

Scherer separated the bile-pigment from urine containing large quantities of it by means of chloride of barium, in the two following ways: he either decomposed the baryta-compound with carbonate of soda, threw down the pigment with hydrochloric acid from the soda-solution, and purified it by solution in alcohol containing ether, by washing with water, &c.; or the baryta-compound was extracted with alcohol containing hydrochloric acid, the solution evaporated, extracted with water, and then treated in the manner above described.

*Tests.*—Unless the amount of bile-pigment in a fluid be not too minute, nitric acid, especially if it contain a little nitrous acid, gives the very characteristic play of colors which we have already described. When, however, the coloring matter is present in small quantity, or when it has already undergone a partial modification, nitric acid often fails to give any appreciable reaction. Schwertfeger's<sup>1</sup> method in such cases is to precipitate the fluid with basic acetate of lead, and to extract the precipitate with alcohol containing sulphuric acid: if any of the pigment be present, the alcohol assumes a green tint. Heller<sup>2</sup> recommends that a little soluble albumen should be added to the fluid to be examined (unless, indeed, it be already albuminous), which must be precipitated by an excess of nitric acid; if any pigment be contained in the fluid, it will communicate a bluish or greenish-blue tint to the coagulated albumen. Heller observes that if ammonia be carefully poured upon urine which contains unchanged bile-pigment, the surface of the fluid assumes a red color.

### *Physiological Relations.*

*Occurrence.*—Bile-pigment usually occurs in fresh bile in a state of solution; often, however, it is in a state of suspension. It almost always constitutes the nuclei of gall-stones; and we sometimes find ramifying nodular concretions in the gall-bladder and in the biliary ducts, consisting almost entirely of bile-pigment. This pigment is found, not only in the bile of man and of the ox, but also in that of other carnivorous and herbivorous animals; it presents, however, the most varied modifications, as we find from the difference of color exhibited by the bile not only of different genera but even of different individuals of the same species; thus, the bile of a dog is of a yellowish-brown tint, that of the ox is brownish-green, while that of birds, fishes, and amphibia is usually of an emerald green.

<sup>1</sup> Jahrb. f. prakt. Pharm. Bd. 9, S. 375.

<sup>2</sup> Arch. f. Chem. u. Mikrosk. Bd. 2, S. 95.

The bile-pigment which mixes with the *contents of the intestines* becomes very rapidly modified, and ceases to present the ordinary reaction with nitric acid; the change which it here very rapidly undergoes, appears to be the same which we can induce artificially by nitric acid. It is in this form that it occurs in the *solid excrements*, unless when diarrhoea is present, in which case unchanged pigment is found in the alvine dejections. It is only rarely that the excrements assume a green tint from the green modification of the pigment; the green coloration more frequently depending on an admixture of partially decomposed blood. Bile-pigment is never entirely absent in the excrements except in the rare cases of icterus, which are accompanied with a complete stoppage of the biliary secretion.

Bile-pigment occurs in the *blood* and in *serous fluids* in all forms of icterus; sometimes however it is absent, or at all events, cannot be detected in the blood in certain forms of inflammation, while cholic acid or its conjugated acids may be recognized; the converse case, namely, the presence of bile-pigment and the absence of cholic acid in the blood is, however, more frequently observed. We shall return to this subject.

In diseases the bile-pigment is especially deposited in the fluids of the cellular tissue, in the aqueous humor, the vitreous humor, the crystalline lens, and above all in the sclerotic; cases have even occurred in which the saliva and the sweat have been colored yellow; sometimes the organism may so long endure this impure condition of the blood, that the pigment saturates even the cartilages, ligaments, and bones,<sup>1</sup> and may actually be recognized in the nerves.

Scherer<sup>2</sup> often discovered decided traces of bile-pigment in the *urine* of healthy persons, especially during the hot months. In disturbances of the function of the liver this pigment very frequently presents itself in the urine, and may usually be recognized by a brownish-red or cinnamon-brown, dark color, which sometimes, if the urine be allowed to stand till it becomes acid (Scherer), passes into a dark-green tint. Sometimes, however, it is also absent in this fluid while other biliary constituents are present in it. Occasionally, in perfect suppression of the biliary secretion—as for instance in true granular liver, when the urine throws down an intense scarlet sediment—no trace either of bile-pigment or of cholic acid can be detected.

*Origin.*—As we are still unable to obtain an empirical formula for the composition of bile-pigment, chemistry affords us no information regarding the origin of this substance. The opinion has certainly long been advanced that bile-pigment was formed from hæmatin, in consequence of the greenish shades of color which extravasated blood usually exhibits, as for instance under the skin after contusions, in the sputa of patients with pneumonia, and sometimes in typhous stools. However plausible this view may appear when we examine the blood-corpuscles of portal blood and find the coloring matter essentially changed in them, yet physiological facts are still wanting to support it. Virchow,<sup>3</sup> by his

<sup>1</sup> Kerkring, Spicil. anat. obs. 57, p. 118.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 181–195.

<sup>3</sup> Arch. f. pathol. Anat. u. s. w. Bd. 1, S. 427–431.

physiological investigations, has with much ingenuity pointed out the way which the chemist must proceed in order to decide the question in reference to this pigment. He was the first to draw attention to the red crystals which are found within the animal organism and which evidently arise from stagnating bile, and to show that in their reactions they take an intermediate place between hæmatoidin and bile-pigment, forming a transition stage between these two pigments.

*Uses.*—Whether the bile-pigment takes any part in the process of digestion, and what are its uses in the intestinal canal, are questions which for the present must remain altogether undecided. The fact that it undergoes so decided an alteration in the intestinal canal leads us teleologically to infer that it fulfils some special object.

These crystals, which are possibly identical with the bilifulvin found by Berzelius in bile which had already undergone change (*Fel tauri inspissatum*), have been found on the wall of echinococcus-sacs, which, in consequence of ruptures and partial resorption of the walls, communicated with the biliary ducts.

The facts now in our possession seem to indicate that the liver is not the part of the organism in which the bile-pigment is formed; we shall, however, discuss this question, when treating generally of the origin of the bile.

## URINE-PIGMENT.

Considered either in a chemical or in a physiological point of view, there is scarcely any substance in the whole range of physiological chemistry regarding which our knowledge is in so unsatisfactory a state as the urine-pigment.

Experiments have often been commenced upon this substance, but the difficulties which present themselves in the investigation are so numerous that most experimentalists have soon resigned it, and directed their labors to some more productive department of chemistry. It unfortunately happens that no certain chemical differences can be detected between urines presenting the most striking difference of color to the eye of the clinical physician.

The difficulties of this investigation are dependent on the following circumstances.

The amount of this substance in the urine is extremely minute; a very small quantity of the pigment giving a color to an extremely large amount of other matters.

It begins to decompose even during the most cautious evaporation of the urine: to be convinced on this point we need only compare urine concentrated by evaporation, with a specimen from which a great part of the water has been removed by congelation.

Even on exposure to the air, or under the air pump, the decomposition of this substance commences.

Like many other pigments, it adheres tenaciously to other substances, sharing their solubility or insolubility.

Besides the pigment, there are other substances in the urine which

have the same degree of solubility, which do not crystallize, and are not volatile; as they neither combine in definite proportions with other bodies, nor differ in solubility from the pigment, they cannot be separated from it.

The pigment occurs in the urine under various modifications, on which are dependent the different tints presented by morbid urine and its sediments.

Finally, this pigment is very readily acted on by chemical reagents, especially by acids and alkalis.

Scherer's<sup>1</sup> investigations on this subject especially show that this pigment is in a state of constant change, that it is decomposed by neutral and basic acetate of lead into two substances, differing in their respective amounts of carbon and hydrogen; and that in a healthy condition of the system it is poorer in these two elements than when there are diseased conditions of the organism impeding the pulmonary or cutaneous transpiration, or the secretion of bile. That portion of the coloring matter which is richest in carbon, forms, as has been found by Scherer and Heller,<sup>2</sup> a dark-blue powder, which when dried, possesses a coppery lustre similar to indigo, and dissolves in alcohol with a splendid purple color. This latter variety of pigment is especially frequent in Bright's disease. Heller distinguishes three such pigments, *uroxanthin*, *uroglau-cin*, and *urrrhodin*.

It is a matter of common experience in science generally, and in chemistry more particularly, that the most circumstantial details are given in reference to the more obscure and less investigated departments, and that deficiencies of knowledge are concealed by an enumeration of unconnected or inaccurately observed facts, or by the most illogical deductions. For ourselves, however, we prefer to confess our ignorance, and to spare our readers from the accumulation of individual features which are incapable of affording a characteristic representation of the subject we would illustrate. Chemists still reckon the urine-pigments amongst what they term extractive matters, and may be said by this arrangement to make a candid avowal of their ignorance in reference to these substances.

Those who may be desirous of attempting to elucidate this obscure subject experimentally, may derive considerable advantage from the study of the older writings of Prout, Berzelius, and Duvernoy, and the more recent memoirs of Heller and Scherer.

## EXTRACTIVE MATTERS.

The above observations on the coloring or extractive matters of the urine, lead us to the consideration of extractive matters in general, and of those of the blood in particular. The term *extractive matter* is ap-

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 180, 195.

<sup>2</sup> Arch. f. Chem. u. Mikrosk. Bd. 2, S. 161, 178.

plied by chemists to those bodies which, whether they are chemically produced, or exist preformed in an animal fluid, exhibit few distinguishing properties (that is to say, are uncrystallizable, incapable of entering into any crystallizable or stoichiometrically constituted combinations with other substances, are not volatile at a certain degree of temperature, &c.), and cannot therefore be separated, or exhibited in a pure state. Modern science has indeed made considerable advance, by learning on the one hand to avoid as far as possible the formation of such substances, and on the other, to separate some of them, and render them more accessible to accurate chemical investigation. We will here observe that substances, such as albuminate of soda, Mulder's binoxide and teroxide of protein, creatine, the inosnates, &c., have been reckoned among the extractive matters; and as many better known substances (as urate of soda, hippurate of soda, and others), are impeded in their crystallization, and are enveloped or concealed as it were by the extractive matters, they also have been embraced under the same head, and have likewise been regarded in the light of extractive matters, and have been calculated as such in analyses. When we consider that the matters circulating in the blood are, on physiological grounds, engaged in an almost constant metamorphosis, we shall easily comprehend the difficulties that beset the chemist in his attempt to seize them at any definite stage of their metamorphosis, especially as they only circulate through the blood in small quantities for the purpose of being deposited in some tissue, or of being eliminated from the organism by the organs of excretion.

The extractive matters must, therefore, be likewise regarded as important factors in the metamorphosis of animal tissue. In accordance with the views of Berzelius, these bodies were considered for the most part as products of the metamorphosis of tissues which, having become unfitted for further purposes, after fulfilling their function, are elaborated in the blood in the better known form of excrementitious matters. But to regard these substances as of a purely excrementitious nature, was taking too circumscribed a view of their importance. Since the blood contains the products of the metamorphosis of the tissues no less than the elements necessary for their formation, it is not only possible but probable that plastic and useful matters, as well as the products of regressive formation, may have been comprehended under the head of extractive matters; for, as we have already observed (p. 37) the idea of the progressive and regressive metamorphosis of matter cannot be followed through an unbroken series of sequences. Albuminate of soda, fibrin itself, and Mulder's protein-oxides, cannot assuredly be regarded in the light of excrementitious substances, but must rather be considered to constitute the transitions from albuminous to gelatigenous substances.

When we reflect that the different stages of metamorphosis of such non-nitrogenous bodies as the fats and carbo-hydrates increase the number of the extractive matters, it seems worthy of notice that their sum in the blood should not be greater than we generally find it to be. But this circumstance proves that very small quantities of the substances which must necessarily occur in the blood, appear simultaneously; and hence the difficulties of the inquiry are considerably increased. The reasons why we are thus unfortunately constrained to continue the use

of the term extractive matters, are sufficiently clear, but yet we cannot refrain from expressing our surprise that, considering the present condition of our science in this respect, chemists can venture to speak of different crases of the blood, or attempt to make them serve as the foundation of a presumed exact humoral pathology.

## NITROGENOUS HISTOGENETIC SUBSTANCES.

The substances belonging to this class present, like the fats and carbohydrates, such great similarities in their composition, and in their most essential properties, that chemists, even if they were unacquainted with their occurrence in the animal body, and with their great physiological importance, would naturally have placed them in one group, seeing that the following properties are common to all of them.

• In the dried state they occur in a solid mass, or in powder, or form gelatinous, brittle, translucent plates; when moist, they are either translucent and yellowish, opaque and white, solid and elastic, soft, tough, and adhesive, or, finally, jelly-like and slippery. All these substances are uncrystallizable, and, unless when an intermixture of other substances is present, are devoid of taste and smell. By far the greater number of them are insoluble in water, and the few which are soluble in it can readily undergo a conversion into a modification insoluble in that fluid; although their physical properties are essentially dependent on and modified by water, and although when dried they condense water with very great rapidity from the atmosphere (and are therefore highly hygroscopic), yet they show little tendency to form definite hydrates, that is to say, chemical combinations with water; they are insoluble in alcohol, ether, and in all neutral menstrua; none of them are volatile: many of them certainly fuse when heated, but not until decomposition has already commenced; at a higher temperature, after the loss of water, they develop a large number of nitrogenous and non-nitrogenous, basic and neutral products, in addition to ammonia, evolving at the same time an unpleasant odor, which is usually compared to that of burnt horn.

A very large number of the substances belonging to this group dissolve unchanged in *acetic* and other organic acids, as well as in common phosphoric acid; and also partially in other mineral acids in a state of extreme dilution. On the other hand, almost all of them are decomposed by *concentrated mineral acids*; many of them swell and assume a gelatinous appearance in *sulphuric* and in *hydrochloric* acid; after prolonged digestion, they form, together with ammoniacal salts, brown humus-like substances, which consist mainly of leucine and tyrosine (see pp. 133–5), and a crystallizable stinking volatile substance, which has not yet been accurately investigated. All, more especially when they are heated, assume a more or less intense yellow color, when treated with *concentrated nitric acid*.



They are all metamorphosed by prolonged *boiling with water*; and the metamorphoses they thus experience from being heated with water, have led to their classification into *albuminous* and *gelatigenous substances*.

The alterations experienced by these bodies from the action of *oxidizing substances*, as for instance, chromic acid or manganese and sulphuric acid, have been most accurately studied during the last few years by Schlieper<sup>1</sup> and Guckelberger;<sup>2</sup> and it is worthy of remark that the non-nitrogenous products of this process of oxidation belong to the butyric acid group, embracing all the acids from formic to caproic acid and their aldehydes; besides these we must also reckon benzoic acid and hydride of benzoyl; but excepting ammonia and hydrocyanic acid, there are only very few nitrogenous products, namely, the nitriles of some of the acids of the butyric acid group.

Some few of these substances are dissolved by the *caustic fixed alkalis* in such a manner, that they can be again precipitated by acids in a perfectly unchanged condition; but the majority can only be dissolved in a concentrated alkaline solution, and with the continued application of heat, by means of which they become perfectly decomposed. Since the greater number of the bodies belonging to this group contain sulphur in addition to the ordinary elements of organic substances, the first effect produced by the action of heated dilute alkaline solutions is the abstraction of the sulphur by the formation of liver of sulphur and of alkaline hyposulphites. There is always a development of ammonia, although this is most considerable when concentrated alkaline solutions are used; carbonic and formic acids volatilize with the ammonia, while new bodies appear in the decoction, having either an acid, or a nitrogenous basic, or indifferent character, as for instance, leucine, glycine, protide, &c. If these substances be mixed with alkalis and gently fused, there will appear a large quantity of cyanide of potassium, leucine, tyrosine, &c., besides the ordinary products of the dry distillation of nitrogenous substances.

It is worthy of remark that these substances have the property of being reduced to the humid condition of *putrefaction* without any apparent or recognizable agency of other matters, and solely by the influence of atmospheric agents. While it is proved that other organic substances admitting of ready decomposition, as, for instance, urea, are not decomposed by the atmosphere even under the most favorable conditions, if they are in a chemically pure condition, the connection of the elementary molecules of these bodies is so easily disturbed by the most ordinary atmospheric influences, that in the presence of water, and at an ordinary temperature, they begin to decompose in the course of a few hours, or, at all events, in a day or two. The period during which they can resist these influences, that is to say, the commencement of decomposition, depends greatly on the state of cohesion in which the molecules occur. The substances deposited in comparatively dense and insoluble masses in the animal tissues, pass far more slowly into a state of putrefaction than the more finely distributed substances, or those which are dissolved in

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 59, S. 1-32.

<sup>2</sup> Ibid. Bd. 64, S. 89-100.

water. The substance of the tendons putrefies less rapidly than cellular tissue and coagulated albumen, and the latter less rapidly than soluble albumen. The products of the putrefaction of these substances have not yet been sufficiently investigated; but among them are always to be found carbonate, butyrate, and valerianate of ammonia, sulphide of ammonium, leucine, and tyrosine.

It is further worthy of observation that all histogenetic substances are *invariably accompanied with fats, alkalies, and salts of lime*, from which it is impossible or very difficult to separate them without decomposition. It is not improbable that in the majority a portion of these admixtures is chemically combined with them; and although but few of these chemical combinations, as that of casein and phosphate of lime, admit of actual demonstration, many chemists are disposed to regard a part of these adhering matters as chemically combined, since the most ordinary indifferent solvents are unable to separate them, while the more powerful agents exert a decomposing or at least a metamorphic action on the main substance; and this applies more especially to the mineral substances accompanying these matters. Rose's investigations<sup>1</sup> regarding the mineral substances, have recently given greater weight to the idea that they may in part at least be combined in a nonoxidized condition with nitrogenous bodies, as has long been conjectured, in accordance with Mulder's views, to be the case with the sulphur, and in part also with the phosphorus of these substances. Rose has advanced very satisfactory grounds for believing that a portion of the alkalies and alkaline earths is contained in these matters in a metallic condition, and combined with radicals containing phosphorus and sulphur. We purpose, however, reverting to this subject under the head of "the mineral substances of the animal body."

It may easily be inferred from the above-named properties, that *it is extremely difficult or perhaps quite impossible to exhibit these bodies in a chemically pure condition.*

By their not crystallizing, and by their not volatilizing without decomposition, we are deprived of two most important means of readily isolating them from other substances; while the readiness with which they are decomposed, has hitherto prevented us from ascertaining which of the above mineral substances are chemically combined, and which are simply mixed with them. This refers specially to the soluble bodies of this class, as albumen, casein, &c., none of which have as yet been exhibited in a chemically pure soluble form. We are still more in doubt in reference to the insoluble substances deposited in the tissues; for even if we succeed (which we rarely can) in extracting from them all mineral substances, we yet have no guarantee that there is only one simple, organic substance deposited in the remaining mass of tissue; and both microscopic and microscopico-chemical investigations have rendered it probable that several chemical substances are mechanically deposited by the side of one another in many of the animal tissues, as quartz, mica, and feldspar, occur together in granite, and cellulose and the incrusting matter, in vegetable cellular tissue. It is often impossible to determine

<sup>1</sup> Ber. d. Akad. d. Wiss. zu Berlin Decbr. 1848, S. 455-462.

whether, after treating animal tissues with the more powerful solvents, the dissolved matter was originally only mixed with the undissolved, or whether it must be regarded as the product of decomposition of a body having a more complicated composition.

We might perhaps succeed in exhibiting these substances in a chemically pure condition, and in acquiring a more accurate knowledge of their chemical constitution, if they could only be united with other substances in definite proportions, and admitted, if possible, of a single neutral combination; but such, unfortunately, in very few instances is the case. Many, it is true, obviously enter into chemical combination with alkalies, with the oxides of heavy metals, and even with acids, but as these combinations are mixed with other bodies and other compounds, we are hindered from establishing by analysis any definite relation between any two of these substances. Moreover, putting out of the question the alkaline and earthy salts that are blended with them, we find that no definite conclusions can be formed from the combinations of such animal matters with oxide of lead; for this oxide (which, with oxide of silver, we prefer to the other metallic oxides, since it almost always forms anhydrous compounds with organic substances, or compounds that can be readily deprived of their water) is found to combine with these bodies in more than one proportion; these compounds are then simultaneously formed, and cannot be separated from one another. The analysis exhibits more or less oxide of lead, according as the neutral compound is mixed with more or less of the basic compound. Hence we can readily understand the cause why chemists have succeeded in so few instances in determining with any certainty the saturating capacity and the atomic weights of these animal substances.

In the *arrangement* of these bodies we are again compelled to have recourse to a physiological principle of classification, which is the more admissible from the circumstance that chemistry here affords us no assistance. Our deficient knowledge regarding the chemical properties of the bodies included in this class, does not enable us to establish a purely chemical basis on which to ground their arrangement. But physiology so far aids us, that it indicates which of these substances are to be regarded as original and protogenic in the animal body, and which are to be regarded as originating from these by a zoo-chemical process, and constituting their derivatives. The protogens or aborigines of these substances, which are, in part, found in the embryo, bear so striking a resemblance to one another, that chemists have discovered only very slight, fluctuating, and often merely relative differences between them. We cannot wonder, therefore, that chemists should have conjectured that these, which had previously been termed *albuminous bodies*, possessed one common radical.

Mulder believed that he had discovered this radical, which, from its great importance, he designated as *protein*, whilst he regarded the ordinary albuminous substances as combinations of this protein with sulphur and phosphorus, or simply with sulphur, and therefore called them *protein-compounds*. Although great doubt has recently been thrown on Mulder's view of protein and its compounds, we yet retain these names for the sake of facilitating our comprehension and general examination

of these combinations. We purpose considering the protein-compounds or albuminous bodies in the first group of histogenetic substances. As, however, physiological chemistry has shown, with great appearance of probability, that all other nitrogenous animal substances are derived from these protein-compounds, we will comprise, under the second group, all those more generally diffused substances of the animal body, which may be regarded as proximate or remote derivatives of these compounds.

#### PROTEIN-COMPOUNDS.

The bodies belonging to this group occur not only in animals, but also to a certain extent in plants. They were for a long time regarded as merely different isomeric modifications of one and the same compound; but subsequently, as already observed, they have been considered by Mulder to be combinations of one and the same atomic group with sulphur and phosphorus. The difficulty of solving this question will be made apparent on comparing the properties of these substances, and considering the observations already made (at pp. 39-40) on the determination of the atomic weights. It must rather excite our surprise that chemists should have hazarded any theory of their composition, than that nothing positive should as yet have been ascertained regarding their composition and mutual relations. Although we have the most accurate analyses of the protein-compounds, it is impossible to form any decisive conclusion regarding their internal constitution; for although the exactness of Mulder's analyses is undoubted, their accuracy must yet be only commensurate with the present comparatively imperfect state of analytical chemistry; that is to say, the empirical results of the analyses of these bodies do not admit of our deciding with scientific certainty on their composition. Hence a formula deduced from these analyses must be simply hypothetical, since several formulæ may frequently be derived with equal correctness from one and the same analysis. In making choice of one of these formulæ we must therefore adopt that which appears to guide us in the best direction, bearing in mind that we have to deal with hypotheses only, and not with facts.

Keeping this consideration in view, we have, in the following remarks, adhered to Mulder's recent hypothesis, in accordance with which albuminous substances are regarded as combinations of a purely hypothetical substance, incapable of being exhibited in an isolated form, with different quantities of sulphamide and phosphamide. We only follow this hypothesis, because from the want of a safer guide, it seems the best adapted to lead us in our advance through this obscure department.

The following properties are common to all the protein-compounds. Most of them occur in two conditions, namely in a soluble and an insoluble or scarcely soluble state; in the former condition, we find them naturally existing in the animal fluids, while they are principally obtained in the latter form by boiling. The soluble modification forms in a dry condition a faint yellow, translucent, friable mass, having no smell or peculiar taste; it dissolves in water, but is insoluble in alcohol and ether: it is precipitated by alcohol from the aqueous solution, after which it is

usually insoluble in water; the aqueous solution may have either a slightly alkaline or a slightly acid reaction, which depends, however, more on the alkali or acid mixed with it than on the substance itself. The aqueous solution is precipitated by most metallic salts, and the precipitate generally contains the acid and base of the salt employed in addition to the protein-compound. The greater number cannot be precipitated from their aqueous solution by alkalies or by most of the vegetable acids, but they are precipitated by mineral acids (with the exception of ordinary phosphoric acid) and by the tannic acids.

Most of them are transformed into their insoluble state by boiling, some by acetic acid, and almost all by the mineral acids; with the latter they usually form compounds soluble in pure water but insoluble in water to which an acid has been added, and incapable of being restored to the soluble modification by saturating the acid with the base. The protein-compounds, when precipitated by salts, usually assume the insoluble form.

The insoluble compounds, when dried, are white and pulverizable; when newly precipitated they are usually of a snow-white color, flocculent or in small clots, or else tough and gelatinous, without taste or smell, without reaction on vegetable colors, and insoluble in water, alcohol, ether; and all indifferent menstrua; they are all more or less readily dissolved by *alkalies*, from which they can be precipitated by mere neutralization with acids. They behave very differently towards different *acids*; they are dissolved by concentrated *acetic acid* and other organic acids, as well as by ordinary phosphoric acid, and are precipitated *from these solutions by yellow as well as red prussiate of potash*. They do not dissolve in moderately concentrated mineral acids, although they combine with them, and these compounds have the property of being insoluble in water to which an acid has been added, although they dissolve in pure water, after having first swelled and assumed a gelatinous appearance. They swell in the same manner in concentrated sulphuric acid, but they assume at the same time a brownish color, and become decomposed. Their relation to *concentrated nitric* and hydrochloric acid is highly characteristic; the former acid giving them when heated a deep *lemon-colored* tint, while *concentrated hydrochloric acid* causes them to assume a gradually increasing intensely *blue* color when exposed to a moderate warmth and to a sufficient supply of air. A fluid obtained by the solution of 1 part of mercury in 2 parts of nitric acid containing  $4\frac{1}{2}$  equivalents of water, forms the most delicate test for the protein-compounds (Millon),<sup>1</sup> whether they are dissolved in a fluid or simply interspersed in a tissue. The fluid, or the tissue that has been moistened with it, is then heated to from 60° to 100°, when an intense red color is observed, which does not disappear either on prolonged boiling or exposure to the atmosphere.

The protein-compounds, when submitted to dry distillation, when allowed to putrefy, and when decomposed by oxidizing agents, behave precisely in the manner of the histogenetic substances generally, which has been already described (pp. 286–287); giving rise to the above-named products of decomposition, although in different relations of quantity.

All protein-compounds contain *sulphur*, which can be very readily

<sup>1</sup> Compt. rend. T. 27, p. 42–44.

detected in these substances both in their natural state, and when boiled, either by heating them with a little alkali on silver foil (when a yellowish-brown spot of sulphide of silver will be formed), or by boiling their alkaline solution for some time with strong acids, when sulphuretted hydrogen will be developed, or with acetate of lead, when sulphide of lead will be precipitated. It is, however, worthy of notice that the protein-compounds may contain sulphur under conditions in which its presence cannot be detected, as Mulder has shown, by the ordinary tests. These were the bodies which were at one time regarded by Mulder as protein, or the non-sulphurous constituents of albuminous matters, but he has subsequently discovered<sup>1</sup> that the substance formerly termed protein contains sulphur. On treating albuminous substances with a dilute solution of potash as prescribed for the preparation of this supposed protein, they lose the property of indicating the presence of sulphur by the ordinary tests. Mulder endeavors to explain this phenomenon by supposing that those compounds which yield a sulphur-reaction, contain sulphur combined with amide, and therefore as *sulphamide*,  $\text{H}_2\text{NS}$ ; and further, that on treating them with potash, 2 atoms of sulphamide, by assimilating 2 atoms of water, are decomposed into ammonia, which escapes, and also into hyposulphurous acid, which combines with the non-sulphurous atomic group to form those compounds which yield no sulphur-reaction on silver-foil. It certainly is true that all these compounds on being digested with caustic fixed alkalies, develop ammonia, and that those yielding the sulphur-reaction contain more nitrogen than those which do not exhibit it. The assumption of the presence of sulphamide in these substances, must, however, still be regarded as a somewhat hazardous hypothesis, in the first place, because we are as yet wholly unacquainted with this sulphamide, whether in an isolated or combined state; secondly, because a combination of hyposulphurous acid with an organic, scarcely basic substance, is as unlooked-for a phenomenon, as it should not be separable by stronger acids from its combination with the protein; and lastly, because the hyposulphites yield a most evident sulphur-reaction when heated with organic substances on silver-foil. Mulder in like manner assumes that the phosphorus contained in albumen, exists in the state of *phosphamide*,  $\text{H}_2\text{NP}$ , a purely hypothetical body, and totally different from Gerhardt's phosphamide, whose amide nature is moreover very doubtful. These are some of the grounds on which we have been led to regard Mulder's view as a mere scientific fiction. By subtracting the elements of hyposulphurous acid from the composition of those albuminous substances which do not yield the sulphur-reaction, and the elements of sulphamide from those yielding such a reaction, Mulder obtained a group of atoms of carbon, hydrogen, nitrogen, and oxygen, which in all these compounds exhibited perfectly identical relations, or only a slight increase of oxygen. This complex atomic group contained in 100 parts 54.7 of carbon, 6.8 of hydrogen, 14.2 of nitrogen, and 24.3 of oxygen. For this complex group Mulder has calculated the formula  $\text{C}_{36}\text{H}_{25}\text{N}_4\text{O}_{10} + 2\text{H}_2\text{O}$ , which expresses, according to him, the true composition of the perfectly non-sulphurous protein.

The sulphur which is not detected by the above-named reactions can

<sup>1</sup> Chem. Untersuch. übers. v. Völcker. H. 2, S. 179-272.

only be discovered and quantitatively determined by the dry method; fusing the dry, organic substance with a mixture of alkaline nitrates and carbonates or caustic alkalis in a silver crucible till the fused mass becomes perfectly white, when the sulphuric acid which has been thus formed, can be determined from the residual saline mass.

Dana<sup>1</sup> has recommended a very good method of detecting the presence of sulphur in organic matters containing that substance in not very minute quantity. We make a mixture of carbonate of soda, starch, and the substance to be tested for sulphur, and heat it by the blow-pipe on a platinum support; we then place the fused mass in a watch-glass with a drop of water, and add a small crystal of the nitroprusside of sodium discovered by Playfair;<sup>2</sup> if sulphur be present, that is to say, if sulphide of sodium be formed, the fluid will assume a splendid purple color; most commonly a red tint first appears, which, assuming a shade of blue, becomes purple, and finally passes into a very deep azure blue, but even this is not persistent, for the fluid at last entirely loses all its color.

Since the termination of Mulder's investigations on the protein-substances, several other views regarding the *constitution* of complex organic bodies have been promulgated. We have to a certain extent given up the older theory of organic radicals (on which Mulder's view is based), and have turned our views towards the establishment of conjugated compounds, salt-like combinations, and the like. The unexpected discoveries of the resolution (or cleavage) into other substances of amygdalin (Liebig and Wöhler), asparagin, salicin, and populin (Piria), the discoveries of the ammonia-alkaloids (Wurtz), and their theoretical constitution (Hoffmann and Kolbe), and finally, the observation that many nitrogenous bodies when decomposed in various ways, yield special volatile alkaloids (Anderson, Rochleder, Wertheim, and others) give a certain support to the view that the protein-substances may have a constitution analogous to that of these complex bodies, and that there may be contained in them several proximate constituents conjugated together, or combined in the manner of salts. Thus, for instance, Wurtz<sup>3</sup> obtained methylamine from casein by treating it with alkalis, and Rochleder<sup>4</sup> by decomposing it with chlorine, and the latter chemist consequently regards methylamine as one of the proximate constituents of casein. This view seems to gain support from the remarkable circumstance that there is an albuminous substance in the blood of carnivorous animals which crystallizes in prisms, while the corresponding substance in the blood of guinea-pigs and rats crystallizes in tetrahedra. This obviously points at combinations of an analogous kind, in which only one different constituent has entered, which, however, is the cause of the difference in the crystalline form of the otherwise perfectly analogous body. Thus, for instance, according to Rochleder's hypothesis, one of these bodies might contain methylamine and the other ethylamine, in combination with the same group of atoms. We are, however, still deficient in the data which are requisite for the further elaboration of such an hypothesis, partly because the

<sup>1</sup> Chemical Gazette. 1851, p. 459.

<sup>2</sup> Philosophical Magazine, 3 Ser. Vol. 36, pp. 197-221, 271-284, and 348-360.

<sup>3</sup> Compt. rend. T. 30, p. 9.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 73, S. 56.

protein-bodies have as yet been little investigated in relation to these views, and partly because their decompositions, in so far as they are yet known, do not enable us to arrive at any definite conclusions on these points.

## ALBUMEN.

### *Chemical Relations.*

*Properties.*—*Albumen*, the principal representative of the protein-compounds, is distinguished amongst these bodies by its occurrence in very different modifications, which are however not to be sought in a different arrangement of the atoms of this substance, that is to say, in a polymerism or metamerism, but depend alone on the substances mixed with it, as alkalies and salts. Hence the albumen of the blood differs in several points of view, not only from that of the hen's egg, and the latter from that of a dove's egg, but it is even found that the albumen of the blood differs in different persons, and that the albumen of the albuminous fluids of the same individual does not exhibit precisely similar reactions. This is one of the causes that has given rise to the various and frequently contradictory statements abounding in chemical literature, in reference to the individual properties of albumen. Albumen obtained indiscriminately from various sources ought therefore, not to be employed for qualitative chemical experiments, but we should first obtain albumen in a state of the greatest possible chemical purity, and we may then ascertain the modifications experienced in its properties and reactions by the admixture of different substances in different proportions; for striking differences are produced in albumen, not merely by the presence of another body, but by the different proportions in which it occurs. Scherer<sup>1</sup> and myself<sup>2</sup> were the first to investigate the properties of albumen in this point of view, but although we may have succeeded in elucidating some few individual points, no perfect and scientifically conclusive results have been attained; and notwithstanding our investigations, experiments have been subsequently made on albumen, containing various admixtures and taken at random from any sources. We shall in this place limit our remarks to the most important and general relations of albumen, lest, by introducing too many details, we should obscure and confuse our general survey. If even slight admixtures are capable of modifying the properties of albumen, we may readily comprehend how much more powerfully they may be affected by chemical changes, even if small, in the grouping or arrangement of the atoms. We know that some kinds of albumen vary in the quantity of sulphur they contain, and others again in their saturating capacity, but these are relations which require further investigation for their complete solution.

We purpose adhering to the old classification, and considering albumen in its soluble and coagulated states.

*Soluble albumen*, dried in the air, forms a pale-yellowish, translucent mass, which may be easily triturated and reduced to a white powder.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 1-65, and Untersuch. zu Pathol. S. 82, ff.

<sup>2</sup> Arch. f. physiol. Heilk. Bd. 1, S. 234.



The specific weight of the albumen of the hen's egg, from which the salts had not been removed, was found by C. Schmidt<sup>1</sup> to be 1.3144; after calculating for the elimination the salts, the density of pure albumen was found to be 1.2617. It becomes positively electric by friction, and is devoid of smell, taste, and reaction on vegetable colors. It swells in water, assuming a gelatinous appearance, does not dissolve freely in pure water, but very readily in water containing chloride of sodium or any alkaline salt. It is insoluble in alcohol and ether.

After being dried *in vacuo*, or at a temperature below 50°, it can be heated to 100° without passing into the insoluble condition; the aqueous solution, however, becomes turbid at 60°, coagulates perfectly at 63°, and separates in flakes at 75°. When excessively diluted, no turbidity can be perceived below 90°, and coagula will only separate after it has been boiled for a considerable time. Panum<sup>2</sup> has contributed many important facts to our knowledge of the albuminous bodies and of their various reactions, and he has done much to correct our views regarding the coagulation of albumen and of similar matters by heat. He has especially shown, by numerous and very careful experiments, the influence exerted by the presence of salts or small quantities of acids on the separation of the protein-bodies at high temperatures. He found, for instance, that as a general rule, the temperature at which precipitation takes place is low in proportion to the amount of salt that has been added, and that the quantity of acid which is requisite to produce a permanent precipitation at the same temperature, is inversely proportional to the quantity of salt that has been added to the solution of albumen. Panum thinks that he is justified, from these and similar experiments, in considering all our previous ideas of coagulation as "confused;" but this conclusion is most distinctly to be drawn from his experiments, namely, that we must very carefully distinguish precipitated albumen from coagulated albumen. It appears, from my observations, that alcohol acts in relation to the precipitation and coagulation of albuminous matters in the same manner as the salts in Panum's experiments. By the gradual addition of alcohol we can depress the coagulating point of the fluid step by step, till we arrive at a point where the albuminous substance is precipitated, although not coagulated; and then, if not soluble in water, it still dissolves in solutions of the neutral salts of the alkalis. As to what actually takes place in coagulation in those cases in which albuminous substances, under the influence of a high temperature, lose many of their other properties simultaneously with their solubility, we are perfectly ignorant, and Panum's experiments have thrown no light on this point.

Albumen may be precipitated from an aqueous solution by diluted alcohol; the precipitate, however, is not coagulated; but when a large quantity of strong alcohol is added, it is converted into the insoluble or coagulated form. It behaves very differently towards ether free from spirit; it is generally asserted that the albumen of the serum of blood is not coagulated, while that of eggs, on the other hand, is coagulated

<sup>1</sup> Ann d. Ch. u. Pharm. Bd. 61, S. 156-167.

<sup>2</sup> Arch. f. Path. Anat. Bd. 4, S. 17.

by ether; but as this observation is not constant, this supposed variation may be dependent on the degree of concentration of the albuminous solution.

Fatty and volatile *oils* neither dissolve nor coagulate albumen. It is coagulated by *creosote* and *aniline*. \*

Albumen is converted into the insoluble state by most acids, but it is not precipitated by the mineral acids (except by tribasic phosphoric acid) unless when they are added in excess. The organic acids, with the exception of the tannic acids, do not precipitate albumen. Panum has also made some very interesting experiments on this point (the effect of acids on albumen), from which it appears that albuminous matters undergo essential changes even by acetic and ordinary phosphoric acids, so that it is not improbable that these acids, acting catalytically, may decompose the albumen into two new bodies. It does not appear from Panum's experiments that these acids enter into a definite combination with the albumen. One of the bodies arising from the action of acetic or phosphoric acid, namely *acid albumen*, is distinguished from the original albumen by its insolubility in concentrated solutions of neutral salts of the alkalies, and by its solubility in water.

*Alkalies* do not precipitate albumen, but they convert it into the insoluble modification.

The greater number of the *metallic salts* precipitate albumen; the precipitate containing either a combination of a *basic* salt with albumen, or a mixture of two compounds, one of which consists of the acid of the salt and albumen, and the other of the base of the salt and albumen. The albumen generally passes into the insoluble state in these combinations.

Albumen is not usually found *isolated* in solution in the normal animal fluids, but in combination with a small proportion of *alkali*, whose quantity does not admit of exact determination on account of the salts which are also mixed with the albumen. In some experiments conducted by myself on the albumen of hens' eggs, I found that 1.58 parts of soda were directly combined with 100 parts of albumen, calculated as devoid of salts. This albumen has a slightly alkaline reaction, is more readily soluble in water than pure albumen, from which it differs mainly in the form in which it coagulates when the aqueous solution is heated (Scherer); for it does not separate in flakes like pure albumen, but forms a white, almost gelatinous mass, or simply gives rise, if the fluid is more or less diluted, to a milky or only whitish opalescent turbidity. The alkaline reaction of the fluid is more strongly marked after boiling, which proves that at least a portion of the alkali must be separated from the albumen on its coagulation. The liberated alkali combines with a small portion of the albumen to form albuminate of soda, which remains dissolved. This albumen, separated by coagulation, passes, however, in part, through the filter, and very soon clogs its pores. On saturating the solution of albuminate of soda with acetic acid, or some other organic acid, it will coagulate on being heated, like pure albumen, into flakes that may be readily collected on the filter. An albuminous solution, after being thus neutralized, is rendered turbid when diluted with a large quantity of water (about twenty times its own volume); a large portion of the

albumen, poor in salts and free from an alkali, being precipitated from the solution.

This phenomenon is dependent upon the circumstance that the albumen, freed from the alkali by acetic acid, is held in solution by the salts, which, however, when strongly diluted, lose their solvent power, and cause the gradual separation of the albumen.

On treating this albuminate of soda with dilute alcohol, there is a precipitation of albumen free from alkali and poor in salts; whilst another portion combined with more alkali, remains in solution and represents the true albuminate of soda, which we are now going to consider. This precipitate dissolves only slightly in pure water, but readily in aqueous saline solutions.

A further addition of alkali to the normal albumen contained in the animal fluids gives rise to an essential difference in its properties. When the solution has been highly concentrated, it yields, on being heated, a translucent jelly, almost insoluble in water, and containing, according to my observations, 4.69 parts of potash or 3.14 of soda to 100 parts of albumen free from salts. On diluting the solution with water, it no longer yields this colorless jelly or any precipitate whatever, on being heated. The albumen, even appears entirely to have lost its coagulability, but such is not the case, for when treated with an excess of alkali, it becomes converted into the coagulated state even without the application of heat; for if the solution be neutralized with some acid that does not ordinarily precipitate albumen (as acetic acid, tartaric acid, or tribasic phosphoric acid), albumen is separated in a coagulated state. The solution of this true *alkaline albuminate* is distinguished by the circumstance that, on boiling, numerous vesicles are formed at the bottom of the vessel, which adhere so tenaciously, as to impart a brown color to this organic substance in process of formation; its surface also becomes covered on evaporation with a transparent film of coagulated albumen (Scherer), which has frequently caused this albuminate of soda in the animal fluids to be mistaken for casein. This alkaline solution yields, however, on boiling, a perfect coagulum in the form of flakes or masses, if any neutral alkaline salt (such as sulphate of soda, chloride of sodium, or hydrochlorate of ammonia), either in the form of a saturated solution, or in the dry state, has been added to it, previously to its being boiled.

*Acids and metallic salts* behave to these alkaline solutions of albumen, nearly in the same way as to those of pure albumen; but the quantity of the metallic salt which is added, often induces modifications, the newly formed albuminates being in some cases soluble, and in others insoluble in an excess of the metallic salt, or of the albuminate of soda. The greater number of these compounds are, however, soluble in alkalies.

On passing a current of *carbonic acid* through a solution of an albuminous body, as, for instance, through the serum of the blood, white of egg dissolved in water, or a solution of the crystalline lens, a greater or lesser portion of the albuminous matter is always separated.

Panum regards this substance as casein, but milk-casein possesses this property in only the slightest degree. Melsens has made this observation on the white of egg, and, on instituting a microscopic investigation

in union with Gluge, observed membranous matters, and hence he gave to this substance the name of "tissu cellulaire artificiel." I have treated all the known protein-bodies with carbonic acid, but never found that the precipitate, when examined under the microscope, presented any peculiarity; it certainly never had the slightest resemblance to any organic substance or to connective tissue. Moreover, Harting<sup>1</sup> has been at the pains of exposing the error into which Gluge and Melsens have fallen.

*Organic acids* added in excess to albuminous solutions, behave in the same manner as alkalies added in excess, causing the albumen to remain dissolved on boiling; if, however, neutral alkaline salts, such as sulphate of soda, chloride of sodium, or hydrochlorate of ammonia, be added to these solutions, the albumen separates on boiling into flakes or clots. Further, these acid solutions on being evaporated are covered with a membrane similar to that which is formed by casein in acid or alkaline milk.

*Coagulated or boiled albumen* possesses all the properties which we have already noticed as exhibited by the insoluble protein-compounds in general. We will, therefore, simply observe that the albumen in its transition from the soluble to the insoluble state, loses a portion of its sulphur; for sulphuretted hydrogen is developed in appreciable quantity: with acids it enters into combinations that are insoluble in water containing acids, but swell and assume a gelatinous form in pure water, before undergoing solution in it. It may be so perfectly combined with caustic alkalies, as to cause their alkaline reaction entirely to disappear. When heated with concentrated hydrochloric acid it dissolves and assumes a blue color, which inclines more to purple than is the case with any other of the protein-compounds. If albumen be boiled for a long time in water, atmospheric air being not excluded, it gradually dissolves, forming a non-gelatinizing fluid, which contains Mulder's<sup>2</sup> teroxide of protein. Finally, albumen when treated with strong oxidizing agents, as, for instance, chromate of potash and sulphuric acid, or binoxide of manganese and sulphuric acid, yields more acetic acid, benzoic acid, and hydride of benzoyl, and less valerianic acid, than the other protein-compounds.

*Composition.*—Albumen, after being coagulated and extracted with water, alcohol, and ether, has been so repeatedly analyzed, that we shall rest satisfied with giving the mean results of five analyses made by Scherer,<sup>3</sup> and subjoining an analysis recently made by Mulder,<sup>4</sup> and regarded by him as the most exact.

	Scherer.	Mulder.	Rüling.
Carbon, . . .	54.883	53.5	53.4
Hydrogen, . . .	7.035	7.0	7.0
Nitrogen, . . .	15.675	15.5	
Oxygen, . . .		22.0	
Sulphur, }	22.865	1.6	
Phosphorus, }		0.4	
	100.000	100.0	

<sup>1</sup> Nederl. Lancet. Sept. 1851.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 47, S. 300, and Bullet. de Néerlande, 1839, p. 404.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 36.

<sup>4</sup> Scheik. Onderz. D. 3, p. 385.

Rüling<sup>1</sup> found in the albumen of the blood-serum (after subtracting the ash, in accordance with the mean of several experiments) 1.325% of sulphur, and in that of hens' eggs, 1.748%, while Mulder found on an average only 1.3% in the former, and 1.6% in the latter. Albumen always retains chloride of sodium with so much tenacity, that it is almost impossible to separate it by washing. The quantity of phosphate of lime which it contains is very remarkable, for, although variable, it usually amounts to about 1.6%. Mulder found from its combination with oxide of lead, that the atomic weight of albumen is 22483.9, while from the oxide of silver compound, he calculated it at 22190.2. For the reasons already advanced (at p. 288), we are as yet unable to establish an empirical formula for albumen; but Mulder calculates, according to the above hypothesis, that the albumen of eggs is composed of 96.2% of protein, 3.2% of sulphamide, and 0.6% of phosphamide; and deduces from these numbers the very hypothetical formula,  $20(C_{36}H_{25}N_4O_{10} \cdot 2HO) + 8H_2NS + H_2NP$ .

*Products of the metamorphosis of albumen.*—The idea has long been entertained that the best method of deducing a formula for the composition of the protein-bodies, is from the study of their products of decomposition, and this view has given rise to that series of splendid investigations which have emanated from the laboratories of Liebig and of Mulder. The discovery of tyrosine by Liebig, and the decomposition of the protein-bodies by oxidizing agents, as illustrated by the investigations of Guckelberger and Schlieper, may be quoted as amongst the results which have sprung from this idea. But none of these investigations have led us to the goal which we had in view, since, for the most part, they only made us acquainted with the more remote products of decomposition. Mulder, however, in his search after a radical, has established several proximate products of metamorphosis, although he was unsuccessful in the attainment of his proposed object. Scherer, who was one of the first to submit the different protein-bodies to careful elementary analysis, instituted further investigations regarding their qualitative analogies and differences, and always sought to trace the proximate forms of metamorphosis of the protein-bodies, both as they occur naturally in healthy or diseased organisms, in special organs, or in the blood, and as they are artificially formed by the action of the less powerful reagents. Although, as yet, we have attained to no certain conclusion, or, indeed, to any conclusion whatever, we believe that this is the only course which can lead us to clearer views. If the discovery of the crystallizability of one of these substances has afforded us the means of obtaining it in a purer state than formerly, the analyses which I have hitherto instituted of the substance of the different crystalline forms have yielded us no definite distinction; hence we can here only refer to those products of the metamorphosis of the protein-bodies which may be considered as proximate products of their decomposition. The first of these which requires notice is albumen-protein.

*Combinations.*—Albumen-protein contains, according to Mulder, 53.7% of carbon, 7.0% of hydrogen, 14.2% of nitrogen, 23.5% of oxygen, and 1.6% of sulphur. He prepares it by dissolving pure coagulated albumen

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 310.

in a solution containing from  $\frac{1}{200}$ th to  $\frac{1}{400}$ th of caustic potash, and exposing it for the space of an hour to a temperature of from  $60^{\circ}$  to  $80^{\circ}$ . The presence of sulphide of potassium in the solution, may then be proved by the ordinary reagents. If we were at once to neutralize the fluid with acetic acid, there would be a danger that the precipitate would contain an admixture of sulphur, since, in addition to the sulphide of potassium, the fluid must also contain hyposulphite of potash, which on the addition of an acid, deposits sulphur, and forms sulphurous acid; this sulphurous acid again, as is well known, yields sulphur with the sulphuretted hydrogen which is developed; hence the fluid must be exposed to the air, and at the same time, frequently stirred till it ceases to yield any further indication of the presence of sulphide of potassium; then, and not till then, we may precipitate the desired body by acetic acid.

When newly precipitated, albumen-protein is of a snow-white color, and in the form of minute flakes; when dried, it assumes a pale yellow tint, is hard and brittle, swells in water into a jelly, but is insoluble in that fluid as well as in all indifferent menstrua, and for the rest behaves like coagulated albumen, with this exception only, that after the treatment with potash, it yields no indication of the presence of sulphur, either with the salts of lead or on silver foil.

*Paralbumen* is an albuminous substance discovered by Scherer,<sup>1</sup> who met with it on several occasions in the contents of ovarian cysts. It is precipitated from the watery solution by alcohol in granular flakes; these, however, again dissolve in water at  $35^{\circ}$  in the course of a few hours, and give the same reactions as the body in its previous state of solution. The aqueous solution is rendered only slightly turbid by boiling, but thick flakes are deposited if acetic acid be then added, although this acid is altogether devoid of action in the cold solution. Nitric acid induces a considerable precipitate in the ordinary solution, while hydrochloric acid, on the other hand, only gives rise to a slight turbidity, even when added in large quantity. Ferrocyanide of potassium, chromic acid, bichloride of mercury, basic acetate of lead, and tannic acid, throw down abundant precipitates.

*Metalbumen* is the name applied by Scherer<sup>2</sup> to another substance which he found in a dropsical fluid. Like the preceding substance, it is also precipitable from its watery solution by alcohol, and is again soluble in water; it is, however, not precipitable by acetic acid or ferrocyanide of potassium; moreover, on boiling the solution after the addition of acetic acid, there is a mere turbidity and no precipitate.

Similar substances have also been found in the urine in morbid states, especially in Bright's disease, and have received various names.

Mialhe and Pressat<sup>3</sup> believe that they have succeeded in tracing albumen through certain successive metamorphoses; they do not, however, base their views on satisfactory chemico-experimental evidence. According to them, *normal physiological albumen* exists in the fluids in a molecular state, and hence, not being actually dissolved, is not amenable

<sup>1</sup> Verhandl. d. phys.-med. Ges. zu Würzburg. Bd. 2, S. 214.

<sup>2</sup> Ibid. Bd. 2, p. 278.

<sup>3</sup> Compt. rend. T. 33, p. 450.

to the laws of endosmosis; it is, moreover, characterized by its coagulability by heat and by the insolubility of the precipitate produced by nitric acid in an excess of the acid. Its first stage of metamorphosis is represented by the *amorphous casein-like albumen* which is produced by the action of the gastric juice; this is endosmotic, but not assimilable, and is imperfectly precipitated by heat and nitric acid; the precipitate induced by the latter is soluble in an excess of the acid. They apply the term *albuminose* to the endosmotic and assimilable substance, which is finally produced by the action of the gastric juice on the albumen. Mialhe<sup>1</sup> maintains (without any additional evidence) that the substance precipitable by alcohol but again soluble in water, which Verceil and Dollfuss<sup>2</sup> found in the normal blood of the ox and called albumen, is identical with this albuminose. Mialhe<sup>3</sup> has, however, the merit, notwithstanding many errors, of being the first closely to study the changes which the albuminous matters undergo during gastric digestion.

The *acid albumen* of Panum which has been already mentioned in p. 296, appears, from his subsequent and more carefully conducted experiments, to be likewise a product of the metamorphosis or cleavage of the protein-body under the action of acids. This substance, which has also been examined, although less accurately, by Melsens, is formed not only from the albumen of the blood and of white of egg, but also from fibrin and other protein-bodies; thus, for instance, I have seen it obtained from the crystallizable protein-substances. According to Panum, the body precipitated by acetic acid from the albuminous solutions saturated with salt, possesses the following properties: when freshly precipitated it forms white flakes, which again dissolve very freely in pure water; they soon, however, lose this solubility on being dried, and especially on being exposed to the air, and likewise on being heated in saline solutions; on the other hand, their solution in water free from salt, is not rendered turbid by the application of heat. We must here notice the remarkable circumstance, that when a comparatively large quantity of salt is in solution with the substance, a comparatively slight heat is required for the separation of the latter, and, conversely, that when less salt is present, a higher temperature is requisite to effect the precipitation. This substance does not exhibit an altogether uniform behavior towards alcohol or towards metallic salts. Panum's analyses of this body, show that neither the acid which is added, nor the salt, exists in it in a state of chemical combination. Sulphur and phosphorus occur in far less quantity than in the original albumen.

Laskowski<sup>4</sup> obtained from albumen, and likewise from fibrin and casein, on treating them with a dilute solution of potash and afterwards with acetic acid, a product which closely resembled these substances, except that it was soluble in alcohol.

*Preparation.*—We have already shown that soluble albumen cannot be obtained perfectly free from mineral constituents. The soluble modification may be obtained in the greatest purity by neutralizing serum or the white of egg dissolved in water with a little acetic acid,

<sup>1</sup> Comp. rend. Vol. 34, p. 745.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 74, S. 218.

<sup>3</sup> Journ. de Pharm. et de Chim. 3 Sér. T. 10, p. 161-167.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 160.

and extracting with from 20 to 30 times the quantity of distilled water, or with dilute spirit. It is, however, usually prepared by evaporating the serum of the blood, or the white of egg in platinum vessels, either *in vacuo* or at a temperature not exceeding  $50^{\circ}$ , pulverizing the yellow residue, and extracting it with ether, and finally with alcohol.

Coagulated albumen is obtained in a perfectly pure state by washing the precipitate yielded on the addition of hydrochloric acid to solutions of white of egg, with dilute hydrochloric acid, in order to remove the salts, and especially the phosphate of lime; by dissolving the hydrochlorate of albumen in pure water, and precipitating it with carbonate of ammonia. The precipitate is then dried, pulverized, and freed from fat by boiling alcohol and ether.

Wurtz<sup>1</sup> obtained a soluble albumen which, however, contained acetic acid, by treating the albumen of hens' eggs with basic acetate of lead, and removing the lead from the albumen by means of carbonic acid and sulphuretted hydrogen. This albumen reddens litmus.

Hruschauer<sup>2</sup> likewise obtained an albumen that reddened litmus by precipitating albumen with sulphuric acid. After being washed for a period of six weeks it reddened litmus; it was, however, free from sulphuric acid.

*Tests.*—The presence of albumen is in general very easily shown, since the coagulability of a fluid by heat is usually regarded as a proof of its presence; but when we consider that several other substances (to be treated of in the sequel) likewise coagulate when boiled, we must not adopt this property of albumen as the sole means of its recognition, since, as has already been noticed, albumen under some relations either does not coagulate, or presents a scarcely perceptible turbidity. We have already indicated the methods by which the presence of albumen may be detected in very acid or very alkaline fluids; we either neutralize the fluid, or we treat it with a strongly saturated solution of hydrochlorate of ammonia, and then boil it. Many methods were formerly recommended for indicating the presence of albumen, especially when occurring only in very small quantities, among which we may particularly notice nitric acid, corrosive sublimate, bi-chromate of potash to which a small quantity of sulphuric acid has been added, and tannic acid; but these methods were only of value when applied in addition to the coagulation test, since the greater number of the protein-compounds are precipitated by them; they are, therefore, only regarded as conclusive when they yield reactions in a fluid in which no other protein-compound but albumen is generally found. Thus, for instance, when urine coagulates on being heated, and is likewise precipitated by nitric acid, corrosive sublimate, chromic acid, and other means, we entertain no doubt of the presence of albumen, although these tests yield the same reactions with most of the other protein-compounds. As, however, all these reagents collectively yield only a relative proof of the presence of albumen, we can trust but little to the evidence afforded by the mere coagulation of a fluid by heating, since animal fluids, as for instance urine, not unfrequently deposit, on heating, a dense, amorphous precipitate, showing no trace of albumen, and consisting only of phosphates. This is often the

<sup>1</sup> Compt. rend. T. 18, p. 700.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 46, S. 348.



case when the urine is very slightly acid, but the precipitate may be distinguished from coagulated albumen by the addition of a mineral acid, which readily dissolves the earths, or by acidulating the urine, before boiling, with a little acetic acid, when no precipitate will any longer be obtained by boiling, if its presence were dependent on the earthy salts of the urine.

In testing animal fluids, and especially those of a pathological nature, we must particularly observe the form in which the albumen coagulates, for on this, as has already been observed, numerous other relations depend; thus, a flocculent coagulum that admitted readily of being collected on the filter, would show that the albumen is not combined with an alkali, and that the latter must have been extracted from it by an acid, since, in the normal state all the albuminous fluids of the body contain albumen in combination with an alkali, and coagulate like milk, or in a white, opaque jelly. Again, if, on evaporation, an animal fluid from which the albumen has previously been removed by boiling, become covered with a thin, colorless membrane, we have no right to conclude, as is so frequently assumed, that casein is present, but simply that the fluid still contains sufficient alkali to prevent the ordinary coagulation of the albumen, and, in short, that although a portion of the albumen may have been removed by boiling, the fluid yet contains the so-called albuminate of soda or potash.

Morbid blood and exudations frequently contain pure albumen that has been dissolved merely by salts; from these fluids the greatest part of the albumen may be precipitated by dilution with large quantities of distilled water, first as a milky turbidity, and finally in flakes, as was first shown by Scherer.

In the determination of albumen it must always be recollected that we are unable to distinguish it from the similar protein-compounds with that scientific accuracy with which we are able to recognize most other organic substances. We may, indeed, indicate the differences presented by the individual reactions in similar substances; but albumen unfortunately occurs in several modifications, sometimes resembling one and sometimes another protein-compound, while neither the determination of the saturating capacity nor the elementary analyses of these bodies present any marked differences. Our determination of the albumen in an animal fluid must therefore at best exhibit only a relative certainty, and this is specially the case where we attempt to discover coagulated albumen; fortunately, however, it rarely or never occurs in this condition in the animal organism; and from what has already been said (at p. 290) in relation to the properties common to the coagulated protein-compounds, it must be apparent that in the present state of science it is useless to attempt drawing distinctions between them. Since the determination of the atomic weight and the elementary analysis are here unable to throw any light on the subject, we might be disposed to take the quantity of sulphur contained in a substance known to be a protein-compound (see p. 291) as a means of ascertaining its identity with coagulated albumen, fibrin, casein, &c., but it unfortunately happens that the quantity of sulphur contained in one and the same body, as for instance in albumen, is not constant. We must for the present relinquish all hope of distin-

guishing from one another the different coagulated protein-compounds of the animal body, and hence it is utterly absurd to inquire whether it be coagulated fibrin or albumen that exists in tubercles or in carcinoma; and yet this is a point which many adherents of the pathologico-anatomical school believe that they have satisfactorily settled without the aid of chemistry.

The method usually recommended for the *quantitative determination* of albumen in the animal fluids is simply to coagulate it by heat, to collect it on a filter, and to dry and weigh it. At the first glance this method seems to be highly practical, but as soon as we attempt to prosecute it, we find our course impeded by unexpected difficulties, unless we would rest content with such deficient and inexact analyses as unfortunately are too common in pathological chemistry. In the first place, it should be observed that the albumen commonly contained in slightly alkaline animal fluids cannot be regarded as capable of being collected on a filter after its coagulation; for while, on the one hand, some portion always passes through the filter in consequence of its gelatinous or milky character, the filter becomes on the other hand so quickly clogged with the coagulated albumen as to preclude the possibility of washing it out; or the fluid passes so slowly through the filter, that the albumen has time to putrefy. Those who suppose that these evils can be remedied by the use of linen or woollen materials as a filter, can have no idea of the degree of exactness required in a chemical analysis; and we cannot refrain from observing that the greater number of analyses of animal albuminous fluids have been conducted in this manner, without any reference being made to these difficulties. Scherer is the only chemist who has directed attention to these obstacles in the way of an exact determination of the albumen, and given instructions regarding the manner in which they may be avoided. In order to determine with exactness the quantity of albumen in a weak alkaline fluid, we must neutralize or slightly acidulate it with dilute acetic acid previously to coagulating it; on the application of heat, the albumen will then coagulate in flakes, and may be both perfectly and rapidly collected on the filter, through which the fluid will pass in a state of perfect clearness. By this method another error incident to the ordinary mode of determining albumen is avoided, for as we have already observed, some alkali is always liberated on boiling any normal albuminous fluid, the fluid exhibiting a stronger alkaline reaction than it did before the boiling. This alkali forms, with a small quantity of albumen, the so-called alkaline albuminate, which, notwithstanding the boiling, remains perfectly dissolved. A portion of albumen must therefore be lost in the ordinary method, even when the coagulated albumen can be collected on a filter, for, as already observed, some of the albumen actually passes through the filter in a dissolved form. Scherer's method entirely obviates this cause of error; care must, however, be taken not to run into an opposite extreme in treating the albumen with too large a quantity of acetic acid, which would equally occasion a loss of the albumen by its solution in that fluid, and its consequent passage through the filter. Hydrochlorate of ammonia may be employed instead of acetic acid, but in this case a longer boiling is requisite, in order completely to precipitate the albumen from the fluid, and to render

it capable of being collected on a filter. It depends entirely on the other steps of the analysis whether acetic acid or carbonate of ammonia be the best suited for the purpose.

Becquerel has recently employed an optical apparatus for the quantitative determination of the albumen in animal fluids, having availed himself of the discovery made by Biot and Bouchardat, that a ray of polarized light is deflected by albumen in the same manner as by sugar. The plane of polarization of the light is turned towards the left; according to Becquerel, the degree of this deviation is proportional to the quantity of albumen that is present: the rotary power is  $37^{\circ} 36'$ ; each minute corresponds to 0.180 of a gramme, and each degree to 10.800 grammes. It would appear from certain counter-experiments made by Becquerel, that this method is very trustworthy.

This is, perhaps, the most fitting place for drawing attention to a point of the greatest importance in the *quantitative analysis of animal fluids*, as well as of organic parts; we allude to the manner of thoroughly drying substances to be weighed. The *thorough drying* of animal substances which are in themselves hygroscopic, or which contain admixtures of protein-compounds, extractive matters, &c., is by no means so easy as that of already dry substances, which, in order to be submitted to elementary analysis, have been exhibited in a perfectly pure state, and have been reduced to a pulverized condition before weighing. It is obvious that desiccation must be effected with the same care as for an analysis with the combustion-tube, if we would not injure the result of the whole analysis; but the circumstance that the substances must here be weighed on filters (whose weight in a dry condition must be predetermined, and which are, moreover, hygroscopic), and that the substances to be weighed cannot be pulverized beforehand, very much increases the difficulty of our forming accurate determinations. Animal substances mostly form horn-like masses on heating, and become covered during desiccation by a crust of dry matter, which is impervious to the water contained in the interior; hence it is frequently impossible to remove all the water contained in such substances without exposing them to a high temperature *in vacuo* and employing sulphuric acid. We must therefore, when it is possible, simultaneously employ high temperatures, air-pumps, and hygroscopic bodies. As analytical chemistry indicates the numerous methods in which these three agents for the removal of water may be employed, we will here simply observe that the two following methods appear to us to constitute the most expeditious means of attaining a perfect desiccation. We either heat a small and convenient sand-bath under the receiver of the air-pump to about  $110^{\circ}$ , and then place upon it the watch-glass or vessel on which the substance to be dried, together with its filter, has already been laid, and then place the sand-bath with the substance under the air-pump over sulphuric acid, and form a vacuum; or we place the substance to be weighed, together with its filter, in a weighed test-glass, which is surrounded by hot sand, and connected with a hand air-pump provided with a chloride of calcium tube, and the air is then abstracted exactly as in the manner directed by Liebig<sup>1</sup> in preparing bodies for elementary analyses. In either case

<sup>1</sup> Handwörterb. d. Chemie. Bd. 1, S. 860.

the desiccation should be continued as long as the substance is found to experience any loss of weight on being weighed. If the air-pump be dispensed with, and the drying be conducted solely by means of heat, as, for instance, by Rammelsberg's<sup>1</sup> or Liebig's<sup>2</sup> admirable air-bath, the temperature must first be raised to 110° or 115°, and the substance then allowed to cool *in vacuo*, for if this precaution were not adopted, the filter and the animal substance would, during their cooling, abstract water from the air, and thus increase in weight. The method proposed by Becquerel and Rodier for weighing substances, while still hot, seems even less to be relied on: for it is well known that by the heating of one of the scales of the balance, the rising current of air renders the substance to be weighed apparently lighter, and analytical chemistry shows us that hygroscopic substances, after being dried at a high temperature, must be cooled in a closed space over sulphuric acid before their weight can be ascertained with certainty. It is therefore here even more necessary than in the preceding method to repeat the process of weighing, until it yield a constant result.

When we consider that all the results of the analysis of organic bodies are entirely dependent on the completeness of the drying process, it is obvious that we can attach very little certainty to many of the published analyses of pathological products. Becquerel and Rodier, who, next to Scherer, have undoubtedly instituted the best analyses of morbid blood, deem it necessary to observe, as something worthy of special notice, that they have devoted the same attention to the quantitative analysis of the blood that is required for an elementary analysis; although we do not see any reason why less exactness is allowable in the far less controllable analyses of animal fluids, than in elementary analyses. In every analysis, but especially in organic analyses, the utmost care is demanded on the part of the experimenter; and where this is not afforded, the labor will result in nothing better than a loss of time and trouble, and a detriment to science. Indeed most of the analyses made in the department of pathological chemistry have been conducted by chemical *dilettanti*, who deluded themselves with the false idea that they were enriching science, and contributing to the establishment of exact medicine by their approximative estimates. It were better for the cause of science, had it never been weighed down by the unprofitable and crude burden of these analyses.

### *Physiological Relations.*

**Occurrence.**—Albumen occurs in all those animal substances which supply the whole body, or individual parts of it, with the materials necessary for nutrition and the renovation of effete matters. Hence albumen is a principal constituent of the blood, the lymph, and chyle, as well as of all serous fluids. It also occurs in the fluids of the cellular tissue, in the white of egg, in the Graafian vesicles, &c. It is especially worthy of notice, however, that it is only in the uncoagulated state that albumen is found in these parts; for, as we have already observed, it would

<sup>1</sup> Anleit. zur quant. min. Analyse. S. 50.    <sup>2</sup> Anleit. zur quant. chem. Analyse. S. 37.

be an impossibility, scientifically considered, to distinguish coagulated albumen from other insoluble protein-compounds in the animal body.

As we purpose in the second volume entering fully into the quantitative relations of the albumen in the *blood*, it will be sufficient here to observe, that the recent investigations of Becquerel and Rodier,<sup>1</sup> with the older ones of Lecanu,<sup>2</sup> Denis,<sup>3</sup> Simon, Nasse, and others, are tolerably agreed in stating that the quantity of albumen in normal blood fluctuates between 6·3 and 7·1%, and in normal blood-serum between 7·9 and 9·8%; Scherer's<sup>4</sup> is undoubtedly the best method that has yet been proposed for the analysis of the blood, which, according to his results, contains in healthy men from 6·3 to 7·0% of albumen. Nasse<sup>5</sup> and Poggiale<sup>6</sup> found on an average less albumen in the blood of most animals than in that of man, the highest quantity being 6·7%. The blood of men appears from the concurrent observations of experimentalists to contain rather less albumen than that of women.

The *chyle* contains less albumen than the blood, but the quantity is variable, as may readily be conjectured from the nature of this fluid; according to Nasse<sup>7</sup> it averages from 3 to 6%.

Marchand and Colberg<sup>8</sup> found only 0·434% of albumen in human *lymph*, while in that of horses Nasse<sup>9</sup> found only 0·391%, including some fibrin, and Schlossberger and Geiger<sup>10</sup> only 0·62%.

The *white of hens' eggs* contains, according to Berzelius,<sup>11</sup> from 12 to 13·8% of albumen.

The *serous fluids* of the animal body, physiological as well as pathological, contain much less albumen than the serum of the blood, as indeed might be inferred *a priori* from their density; they are however never wholly free from it.

The *animal tissues* are almost all surrounded by albuminous fluid; but the large quantity of albumen found in many of these tissues depends upon the numerous capillaries by which they are intersected; as we specially observe in such organs as the liver, kidneys, brain, and muscles.

In the normal condition no albumen seems to pass into the *secretions*, as for instance the saliva, gastric juice, bile, mucus, &c., for although they do indeed exhibit traces of protein-compounds, these latter differ from ordinary albumen. The pancreatic juice contains, however, in its normal state a substance extremely similar to albumen, which coagulates on being heated, and perfectly solidifies the fluid (as in the white of hens' eggs). This substance may, however, occur in any of these fluids in morbid conditions of the secreting organ; and Jul. Vogel<sup>12</sup> has especially shown that the mucous membranes may secrete albumen in addition to the ordinary mucus-corpuscles, when abnormally excited; (hence the presence of albumen in a fluid resembling pus is no evidence of the presence of true pus, or rather of a suppurating surface.)

Bernard<sup>13</sup> found that the albuminous substance of the pancreatic juice

<sup>1</sup> Gaz. méd. 3 Ser. T. 1, p. 503, &c.    <sup>2</sup> Etudes chim. sur le sang hum. Paris, 1837.

<sup>3</sup> Arch. gen. de Méd. 3 Sér. T. 1, p. 171.

<sup>4</sup> Haeser's Archiv. Bd. 10, S. 191.

<sup>5</sup> Journ. f. pr. Chem. Bd. 28, S. 146.

<sup>6</sup> Compt. rend. T. 25, pp. 198-201.

<sup>7</sup> Handwörterb. d. Physiol. Bd. 1, S. 233.

<sup>8</sup> Pogg. Ann. Bd. 43, S. 625-628.

<sup>9</sup> Simon's Beitr. z. phys. u. pathol. Chem. Bd. 1, S. 449-455.

<sup>10</sup> Arch. f. physiol. Heilk. Bd. 5, S. 391-396.    <sup>11</sup> Lehrb. d. Chem. Bd. 9, S. 650.

<sup>12</sup> Untersuch. üb. Eiter, Eiterung u. s. w. Erlangen. S. 75.

<sup>13</sup> Arch. gén. de Méd. 4 Sér. T. 19, p. 68.

exhibited the same behavior in reference to acids, metallic salts, and to heat, as ordinary albumen, and that it was not coagulated by acetic or lactic acid. Bernard instances as a characteristic difference, that the substance of the pancreatic juice is soluble in water after its precipitation by alcohol, but this, as we have already observed, is likewise the case with albumen when dilute alcohol is used. Concretions taken from the pancreatic duct, and for which I am indebted to the kindness of Professor Hasse, dissolved almost entirely in water and exhibited the ordinary reactions of albumen.

Mack,<sup>1</sup> Vogt, and Scherer,<sup>2</sup> have found albumen in the *liquor amnii*, and the two latter inquirers ascertained from their observations that the amniotic fluid is richer in albumen in the earlier than in the later periods of foetal life.

Vogt found in a fluid of a foetus at the fourth month 10·77%, and in that of one at the sixth month 6·67% albumen. Scherer, however, in that of one at the fifth month, found 7·67%, and only 0·82% in the fluid at the ordinary period of delivery.

In the physiological or normal condition no albumen is contained in the *excretions*, and its appearance indicates either disease of the excreting organ or a complete alteration in the composition of the blood.

The occurrence of albumen in the *urine* may be coincident with very different pathological conditions, although its presence was formerly made to constitute a special disease. Simon even asserts that he has often found albumen in the urine of persons, at all events, apparently healthy. In many acute and chronic diseases, unconnected with affections of the kidneys, albumen not unfrequently appears for a short time in the urine, as, for instance, in inflammation of the thoracic organs, acute articular rheumatism, intermittent fevers, typhus, measles, cholera, insufficiency of the valves or contraction of the orifices of the heart, also in chronic affections of the liver, and in pulmonary and peritoneal tuberculosis, especially towards the fatal termination of these diseases. The transitory passage of albumen into the urine appears to depend in these conditions on a change in the character of the blood, in consequence of which the albumen is able to penetrate through the tissue of the kidneys. It is, however, in affections of the kidneys, whether acute or chronic, that albumen appears most constantly in the urine. Bright's disease is, as is well known, a term of very wide significance, but if we limit it as much as possible, and merely include under the term a degeneration of the tissue of the kidney, more especially of the cortical substance, whether of a fatty or other character, we may regard the presence of albumen in the urine as a constant symptom of this disease. But in transitory renal catarrh, such, for instance, as occurs in erysipelas nearly as frequently as after scarlatina, albumen, together with the well-known epithelial cylinders of Bellini's ducts, is found as constantly in the urine as in inflammatory affections of the kidneys, where it is associated with the fibrinous plugs from the same ducts, and as in true Bright's disease. It is almost unnecessary to observe that the presence of pus or blood in the urine necessitates that of albumen, but it is

<sup>1</sup> Heller's Arch. f. Chem. u. Mikrosk. Bd. 2, S. 218.

<sup>2</sup> Zeitschr. f. wissenschaftl. Zool. Bd. 1, S. 88-92.

worthy of notice that a little albumen, together with mucus-corpuscles, is always found in uncomplicated severe catarrhs of the mucous membrane of the bladder.

The observations already made in reference to the occurrence of albumen in the urine apply almost equally to its appearance in the *solid excrements*. Albumen is always found in the excrements in diarrhoea depending upon intestinal catarrh, and in diseases complicated with this affection; the quantity of the albumen increases, moreover, in proportion to the degree in which the blood becomes altered during the diarrhoea; hence, we find that not only in dysentery and cholera, in which so much stress has been laid on the discharge of albumen, but also sometimes in Bright's disease, albumen, together with entire patches of cylindrical epithelium (in some cases the entire thimble-like coverings of the intestinal villi) is discharged in masses by the rectum.

*Origin.*—We have at present very little definite knowledge regarding the origin of albumen from the nitrogenous food. No doubt can be entertained that the chief source of the albumen of the blood is to be sought in the protein-compounds contained in the food; for independently of the circumstance that direct experiments prove that animals cannot exist on food containing no protein-compounds, we find from comparative statistics of the food which has been taken, and of the nitrogenous matters expended in the metamorphosis of tissue (see "Nutrition" in the second volume), that the animal organism derives more than a sufficient supply of protein-compound from the ordinary vegetable food. Although we are not yet able to decide with absolute certainty on the incapability of the animal organism to generate albumen from other sources than protein-compounds, it yet appears highly probable that such is the case. We are not even acquainted with the mode of origin of the albumen of the blood from the allied protein-compounds contained in the food, as casein, vitellin, fibrin, legumin, &c.: all we know is that these bodies are converted by the process of digestion into substances differing very much in their physical properties from the above protein-compounds, but resembling one another in their solubility in water, their insolubility in alcohol, and their incapability of coagulating. How and where these peptones become converted into the normal albumen of the blood, are points on which we are entirely ignorant, neither can we understand by what process the albumen acquires its due quantity of sulphur, since these peptones, as I have convinced myself, for the most part contain exactly as much sulphur as the substances from which they originate.

*Uses.*—After what has been said of the occurrence of albumen, it seems scarcely necessary to adduce any further proof of its utility in forming and renovating the nitrogenous tissues of the animal body. In fact the whole theory of nutrition rests on this postulate. It is a question that has been much contested and variously answered, whether albumen directly co-operates in the formation of cells and the elements of tissues. Jul. Vogel<sup>1</sup> is an especial supporter of the view that fibri-

<sup>1</sup> Path. Anat. S. 80 ff. [or p. 107, &c., of the English Translation. Vogel's opinion is not quite fairly stated in the text. His remarks apply solely to morbid development.—G. E. D.]

nous exudations are alone adapted for the formation of cells and tissues; basing his opinion on pathologico-anatomical experiments on exudations, and on the fact that a small quantity of fibrin is contained in the lymph for the reproduction of effete materials. The absence of fibrin in the fluids of the egg, must also be considered as opposing Vogel's view, since these fluids exhibit the highest degree of plasticity; yet it must be admitted on the other hand, that this counter-proof is less worthy of attention from the circumstance that vitellin, which is the true germ of the egg, has been found by the most careful investigation to be more similar to fibrin than any other protein-compound, having, indeed, an almost perfectly identical composition with it. But independently of the peculiar relations of the germ of the egg, a careful consideration of plastic exudations will in itself lead us to doubt the correctness of Vogel's view, for how can the small quantity of fibrin in the plasma (see "Fibrin") give rise to the frequently large accumulations of fibrinous exudations that are passing into an organized condition, rapidly as the resorption of the serous portions of these exudations may be effected? We cannot suppose that Vogel intends to assert that it is only the fibrin of the exudations which is converted into cells and fibres. The following mode of considering the subject appears to correspond most closely with the facts before us. We shall in a subsequent part of the work enter upon the consideration of fibrin, as a link or transition stage in the metamorphosis of nitrogenous matter; we agree therefore so far with Vogel as to assume that all albumen passes through a transition stage, which we term fibrin, before it can be converted into cells and the elements of tissues: hence this intermediate link in the metamorphosis of tissue appears in very small quantity or not at all, because at this stage the metamorphosis is stationary for only a short time. If we regard fibrin as a body whose specific gravity is ever changing with its chemical changes, as, for instance, is the case with the aldehydes, it would scarcely remain for any appreciable length of time at a given stage of metamorphism, and would therefore be as little appreciable to our senses as the aldehyde of acetic acid, in the process of acid fermentation. We therefore believe that in the organization of the exudations, fibrin is formed from the albumen of the transuded plasma, but that it rapidly undergoes further metamorphosis.

It still remains, however, for us to determine why cells and fibres are not formed from serous exudations, that is to say, from albuminous solutions containing no fibrin. This question might perhaps be answered by supposing that the presence of fibrin is only required to form the point of crystallization for the deposition of plastic matter, and this view seems to derive support from the fact, that a portion of coagulated fibrin when thrown into an uncoagulated plasma, perceptibly accelerates the coagulation of the fibrin; but so simple an explanation is probably not admissible, and it would rather seem that the serous exudations possess no tendency to become organized, in consequence of their never being pure plasma *minus* fibrin, but of their frequently containing less albumen, and in all cases more salts and extractive matters, than the serum of the blood; although we are unable to determine the manner in which salts are able to arrest the metamorphosis of albumen into cells, we yet



know that other metamorphoses of albumen, as, for instance, putrefaction, are hindered or modified by the agency of these bodies.

## FIBRIN.

*Chemical Relations.*

*Properties.*—We must distinguish between numerous modifications of fibrin, if we would attempt to specify the various substances to which this term has been applied. We purpose, therefore, only to consider fibrin, in the first place, in its naturally dissolved form; next, in a spontaneous state of coagulation; and, lastly, when it is coagulated by heat, or boiled.

In the *natural solution of fibrin*, we can distinguish only a few of its properties, since it is here mixed with albumen and other matters of the serum of the blood; and we are acquainted with few reagents by which to distinguish dissolved fibrin in filtered frogs' blood (*i. e.* in blood deprived of its corpuscles), from the albumen contained with it in solution. We at present know nothing more of dissolved fibrin than the facts long ago advanced by Joh. Müller.<sup>1</sup> Neither acetic acid or caustic ammonia induces a precipitate in the fluid of frogs' blood; but a concentrated solution of caustic potash will precipitate fibrin as well as albumen (see p. 297); ether causes fibrin to coagulate, while it allows the albumen of frogs' blood to remain dissolved. The spontaneous coagulation of the fibrin from the plasma of all vertebrate animals may be greatly retarded by dilute solutions of the alkaline sulphates, nitrates, hydrochlorates, carbonates, and acetates, and may even be entirely prevented by concentrated solutions.

As we purpose treating somewhat fully of the spontaneous coagulation of fibrin when we enter upon the consideration of the blood, we will now merely observe, that the *liquor sanguinis* (after the removal of the blood-corpuscles) will frequently assume a thick fluid and gelatinous character within two minutes after its removal from the living body; in a short time some drops of fluid appear on the tolerably consistent jelly, and speedily augment, until they form an entire stratum of serum over the now fully developed coagulum; this coagulum now begins to contract, becoming more or less tenacious, tough, elastic; and resistant, according to certain accompanying conditions (as we shall more fully explain when treating of the blood). If we trace this transition of the fibrin from the dissolved fluid condition into the solid state under the microscope, a careful observation shows us that the fresh *liquor sanguinis* exhibits nothing morphological beyond some few colorless blood-corpuscles; when it begins to gelatinize, separate points or molecular granules appear at various spots, from which arise extremely fine straight threads, in radiating lines, although they do not form star-like masses as in crystallization; these threads becoming elongated cross those springing from other solid points until the

<sup>1</sup> Lehrb. d. Phys. Bd. 1, S. 117 [or vol. 1, p. 124, of the second edition of the English Translation].

whole field of view appears as if it were covered with a delicate, but somewhat irregular cobweb. This network finally becomes so dense that the colorless blood-corpuscles imbedded in it can scarcely be distinguished.

During the last few years it has been repeatedly maintained, that fibrin does not coagulate in threads, but in lamellæ. We may readily convince ourselves of the accuracy of the description given in the text, if we will take the trouble to use in the experiment inflammatory blood, in which the red corpuscles sink rapidly and the fibrin coagulates slowly. Funke has shown in his Atlas the coagulation of the fibrin from a fresh drop of blood, according to E. H. Weber's method (see figure under head of Blood); but here, on the one hand, the red corpuscles disturb the accuracy of the observation, and, on the other hand, we might regard the coagulation of the fibrin as the separation of a laminated mass, which readily forms plaits or folds on which the fibrillated appearance depends. The off-shooting of individual threads from the molecular granules which first become apparent, the projection of these threads, and their gradual augmentation in various directions, can only be seen in blood with a buffy coat. I am, however, unable to decide whether, at the final separation of all the fibrin, these filaments subsequently increase in two dimensions, that is to say, both increase in thickness and become converted into lamellæ or solid masses; if we observe dried blood, after the addition of water, with the microscope (as, for instance, the thin section presented by a small spot of blood, such as is often presented to us in medico-legal investigations), we see that everything dissolves or disappears except the so-called lymph-corpuscles and the fibrin; under these circumstances, in consequence, doubtless, of the thinness of the section, the fibrin certainly appears in the form of pure lamellæ, in which only a few distinct duplicatures are visible.

It is scarcely necessary, at the present day, to offer any refutation of the older views, according to which, on the one hand, fibrin arose from the bursting of the colorless or even of the red blood-corpuscles, while, on the other, it was simply deposited from the blood in which it was originally only suspended. The former view has long ceased to be held by physiologists, while microscopic observations afford ample evidence of the untenability of the latter hypothesis.

As yet no satisfactory solution has been afforded to the question which has been frequently raised regarding the means by which the fibrin is held in solution in the circulating blood, and by which it is disposed to coagulate on the removal of the plasma from the living body. Various facts prove, indeed, that the access of the air (that is to say, of the oxygen), greatly influences the coagulation of the fibrin; but it is doubtful whether this is the only cause of coagulation, since the same process goes on within the vessels of the living organism, as soon as the blood ceases to circulate. This question cannot be answered chemically, since we are at present acquainted only with the product of this process, while it is requisite for a correct judgment of it that we should know not only the end, but the beginning, that is to say, the substance originally held in solution in the blood. We must, therefore, still limit ourselves to the assertion that the blood of vertebrate animals holds a substance in

solution, which, by its metamorphosis, generates a substance not soluble in the serum of the blood, and which we call fibrin.

The view that formerly prevailed, namely, that the fibrin was held in solution in the blood by alkalies and alkaline salts, and that its coagulation was owing to the decomposition of the combination of the fibrin and the alkali by the carbonic acid of the air, has been thoroughly refuted by Nasse;<sup>1</sup> indeed, blood containing much carbonic acid coagulates very slowly, and on the other hand, the carbonated alkalies retard, and may even wholly prevent the coagulation of the fibrin. If, therefore, we are determined upon seeking an explanation of this phenomenon, we must rest satisfied with mere fiction based upon analogy. Thus we may conceive that the albumen of the blood, while undergoing a process of metamorphosis, is disposed to assume a metamorphosed and insoluble form by the agency of the minutest quantity of oxygen, in the same manner as the juice of the grape, according to Gay Lussac's experiments, is brought into a condition of vinous fermentation by means of the minutest quantity of oxygen. But when so distinguished an inquirer as Nasse, while he declares this process to be a chemical one, regards the substance that undergoes the metamorphosis, as endowed with vitality, we are bound to reject his explanation as mere fiction; for, independently of the fact that if a process be chemical it must be capable of chemical explanation, it seems to us wholly at variance with all preconceived ideas of life to attribute life to a simple organic substance.

*Spontaneously coagulated fibrin* is a yellowish, opaque, fibrous mass, which becomes hard and brittle on drying; it is without smell or taste, and is insoluble in water, alcohol, and ether; after being dried it merely swells in water, and becomes again soft and flexible; it readily decomposes peroxide of hydrogen; it dissolves more easily in acetic acid and alkalies than many other protein-compounds; it decomposes rapidly and putrefies in the air, dissolving, if sufficient water be present, and becoming converted into a substance which, like albumen, is coagulable by heat; during this process it attracts a considerable quantity of oxygen, gradually develops ammonia, carbonic acid, butyric acid, and sulphuretted hydrogen, and leaves a residue consisting principally of leucine and tyrosine (Scherer,<sup>2</sup> Marchand,<sup>3</sup> Wurtz,<sup>4</sup> Bopp<sup>5</sup>). It is generally supposed that spontaneously coagulated fibrin will dissolve in solutions of certain alkaline salts; but we should greatly err if we were to regard a fluid thus obtained as a simple solution; for fibrin not only requires a longer period to dissolve in a saline fluid than is necessary for the solution of a simple substance in an indifferent menstruum, but also a higher temperature, and the saline fluid must always be kept for one or more hours at a temperature approximating to the hatching heat (between 30° and 40°), before any considerable quantity of fibrin will be dissolved. Moreover, the fibrin should not be too long exposed to the action of the air, if we wish to effect its solution. Denis,<sup>6</sup> who first noticed this solubility of fibrin, Scherer,<sup>7</sup> and Polli,<sup>8</sup> used for this purpose a solution of 3 parts of

<sup>1</sup> Handwörterb. d. Physiol. Bd. 1, S. 109 ff.

<sup>2</sup> Lehg. d. physiol. Chem. S. 69.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 69, S. 16-37.

<sup>7</sup> Op. cit.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 35. \*

<sup>4</sup> Ann. de Chim. et de Phys. T. 11, p. 258.

<sup>6</sup> Arch. gén. de Méd. 3 Sér. T. 1, p. 171.

<sup>8</sup> Ann. univ. di med. 1839. Apr. pp. 25-33.

nitrate of potash in 50 parts of water. Zimmermann<sup>1</sup> has however shown that solutions of the alkaline sulphates, phosphates, carbonates, and acetates, as well as the chlorides, bromides and iodides, might be employed for the same object. The solution thus obtained, which is always imperfect, and contains undissolved portions requiring to be removed, is viscid, and at about 73° coagulates in flakes. It differs from an albuminous solution in being strongly precipitated by acetic acid (which is only the case to a slight degree with albumen when carefully neutralized); it is not coagulated by ether, in which respect it differs from the naturally dissolved substance which forms fibrin. When the fibrin has been digested for a sufficient length of time, the solution is not rendered turbid by dilution with water, as is the case after digestion for only a short period. At an ordinary temperature, the clear solution remains for a long time unaffected by the atmosphere, only depositing solid particles after it has absorbed oxygen, when it has passed into a state of putrefaction, and exhibits *vibriones*.

Scherer thought that he had proved that the fibrin from arterial blood or from venous blood in inflammatory diseases could not be converted into this albuminous substance by saline solutions. This view has been contradicted by Zimmermann, but the subject has not yet been fully investigated. My own experiments tend to show that the fibrin of the venous blood of the ox very speedily loses these properties, while that of the arterial blood of the same animal does not dissolve in a solution of nitrate of potash. In man I found that fibrin, whether from venous, arterial, or inflammatory blood, was soluble, excepting in two cases of inflammatory blood; the arterial and venous fibrin from pigs' blood dissolved equally well, and with great rapidity in water containing nitrate of potash.

*Boiled fibrin* possesses almost all the properties common to coagulated albumen, from which it is extremely difficult to distinguish it. C. Schmidt<sup>2</sup> found the specific weight of dry fibrin extracted with water, alcohol, and ether to be = 1.2678 after deducting the influence of the ash-constituents. The influence of heat deprives this fibrin of the property of decomposing peroxide of hydrogen, and of being converted into a soluble, albumen-like substance by digestion in solutions of alkaline salts. With acids and alkalies it reacts in the same manner as coagulated albumen; it dissolves in alkalies, and forms with them compounds having no reaction on vegetable colors; with acids it also forms combinations which are insoluble in water to which an acid has been added, but dissolve freely in pure water. Concentrated hydrochloric acid communicates an indigo-blue color to it. By prolonged boiling in water, it becomes decomposed into a soluble and an insoluble compound, to the former of which Mulder<sup>3</sup> has given the name of teroxide, and to the latter, binoxide of protein. When decomposed by chromic acid, or by peroxide of manganese and sulphuric acid, it yields a larger quantity of butyric acid than any of the other protein-compounds or their derivatives; it yields, however, less acetic and benzoic acid than albumen, although more than gelatin (Guckelberger).<sup>4</sup>

<sup>1</sup> Casper's Wochenschr. No. 30, 1843.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 156-167.

<sup>3</sup> Ibid. Bd. 47, S. 300-328.

<sup>4</sup> Op. cit.

*Composition.*—Before we can consider the chemical constitution of a body, it is always necessary to inquire whether we have to deal with a pure and simple substance, with a chemical compound, or, as is often the case, with a body with which several substances are mixed. The question is more imperative in reference to fibrin than to any other animal substance, for both microscopico-mechanical investigations and many chemical experiments seem to indicate that the ordinary, so-called purified fibrin is not a chemically simple substance. Whether fibrin be separated from the blood or from the lymph, it is invariably found to be mixed with heterogeneous morphological elements, especially with the colorless blood-corpuscles, and what are termed the fibrin-disks, which are found associated with molecular granules of various kinds, and usually even with blood-pigment. A microscopic examination of coagulated and perfectly washed fibrin will readily prove that the mass under consideration is not of a homogeneous nature. It is a chemical fact that pure fibrin (even that of the pig, which dissolves so readily in a saline solution), is incapable of complete solution, and always leaves a quantity of insoluble flakes. Even if Bourchardat's<sup>1</sup> statement is erroneous, as asserted by Dumas, Cahour,<sup>2</sup> and Mulder,<sup>3</sup> that he has decomposed fibrin into epidermose and albuminose, Mulder's experiments undoubtedly tend to show that more than one substance must lie concealed in fibrin; and this seems further proved by the above-mentioned difference in the fibrin in different classes of animals, as well as by its different character in diseases (the molecular fibrin of Zimmermann,<sup>4</sup> the parafibrin and bradyfibrin of Polli).<sup>5</sup> Microscopical examination furnishes us, however, with the chief proof that fibrin is not a simple body.

In considering the elementary composition of this body, we must therefore always bear in mind that the results of the analyses refer to a mixed substance.

We will therefore content ourselves with giving the results of Scherer's and Mulder's analyses, in order to present some idea of the proportion of the various elements constituting fibrin.

	Scherer.	Mulder.
Carbon, . . . . .	53.571	52.7
Hydrogen, . . . . .	6.895	6.9
Nitrogen, . . . . .	15.720	15.4
Oxygen, . . . . .		23.5
Sulphur, . . . . .	22.814	1.2
Phosphorus, . . . . .		0.3
	100.000	100.0

Rüling<sup>6</sup> found 1.319% of sulphur in the fibrin of the blood of the ox, while Verdeil<sup>6</sup> gave it as 1.593%. Most of the later elementary analyses of fibrin agree in the view that there is rather a larger quantity of oxygen contained in it than in albumen; Mulder therefore regards it as a higher stage of oxidation of his hypothetical protein, combined with sulphamide and phosphamide, and assigns to it the hypothetical formula,  $(C_{36}H_{25}N_4$

<sup>1</sup> Compt. rend. T. 14, p. 962.

<sup>2</sup> Ibid. p. 995.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 47, S. 303–305.

<sup>4</sup> Zur Analysis und Synthesis der pseudoplast. Processe. Berlin. 1844. S. 110 ff.

<sup>5</sup> Gazeta med. di Milano, 1844, p. 118.

<sup>6</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 312 u. 318.

$O_{11}.2HO) + H_2NS + H_2NP$ . Fats are always associated with fibrin; and although they have not been thoroughly investigated, they would appear to consist principally of soaps of ammonia and lime. (Berzelius,<sup>1</sup> Virchow)<sup>2</sup>. Dry fibrin contains about 2.6% of these fats.

Like all protein-compounds, fibrin contains mineral substances, of which the principal is phosphate of lime. Mulder found 1.7%, but Virchow only 0.66% of this salt, mixed with a little carbonate of lime.

*Compounds.*—*Fibrin-protein, binoxide of protein*, corresponding, according to Mulder's hypothesis, to the formula  $6(C_{36}H_{25}N_4O_{11}.2HO) + S_2O_2$ , occurs, as we learn from the same observer,<sup>3</sup> in most animal fluids, associated in larger or smaller quantity with fibrin. It may be obtained from boiled fibrin or vitellin, precisely in the same manner as albumen-protein from albumen, or by boiling the fibrin for a long time in water exposed to the air, or lastly by treating hair or horn with a solution of potash, filtering the boiled fluid, and precipitating with acetic or hydrochloric acid. It may be purified by repeatedly dissolving it in caustic potash, and precipitating it with acetic acid.

This body forms a light-yellow, lumpy, tough precipitate, which, when dried in the air, cakes together into a blackish-green, shining, resinous mass, and on trituration, forms a dark-yellow powder; it becomes very viscid in warm water, and admits of being drawn into long, silky, shining bands and threads; it renders water in which it is boiled only slightly turbid, and is perfectly insoluble in alcohol and ether; it dissolves in dilute acetic acid and in dilute mineral acids; nitric acid does not communicate to it so well-marked a yellow color as to the other albuminous substances; when dissolved in acids it may be precipitated by yellow and red prussiate of potash, by tannic acid, and by acetate of lead; it is readily soluble in alkalis, from which it may again be precipitated by acids, it fuses on being heated, and finally carbonizes, with the evolution of a horn-like odor.

*Preparation.*—The method first adopted by Joh. Müller is generally employed for obtaining the natural solution of the fibrin-yielding substance, viz., diluting frogs' blood with sugared water (1 part of sugar to 200 of water), and filtering it.

The best means of obtaining frogs' blood for this experiment is to amputate both thighs, and allow the blood, with which a considerable quantity of lymph is mixed, to flow into sugared water, which not only dilutes the *liquor sanguinis*, but retards the coagulation of the fibrin; the blood-corpuscles of the frog, like those of most of the other amphibia, are, as is well known, much larger than those of mammalia and birds, and therefore pass less easily through the filter.

A considerable quantity of the natural solution of fibrin may be obtained from human blood (the corpuscles of which have the property of sinking very rapidly), by pouring off the very slowly coagulating fluid which collects above the blood-corpuscles.

A single drop of fresh blood, when laid on the object stage and covered with a piece of glass, is sufficient to exhibit the coagulation of the fibrin under the microscope: on account of the mass of red corpuscles the

<sup>1</sup> Lehrb. d. Chem. Bd. 9, S. 88.

<sup>2</sup> Zeitschr. f. rat. Med. Bd. 4, S. 269 ff.

<sup>3</sup> Untersuch. übers. v. Volcker. H. 2, S. 253.

coagulation is however not so well seen as when we employ a drop of fluid from the surface of blood, in which the red corpuscles have sunk below the upper level.

In preparing spontaneously coagulated and boiled fibrin, the blood-clot must be cut into fine pieces, and then washed in water until it appears perfectly white. The fibrin obtained in this manner is more readily washed than when obtained from whipped blood. The process of whipping consists either in shaking the blood, as it flows from the veins, in a bottle with shot, or rapidly stirring it with small twigs or rods; the blood-corpuscles remain suspended in the serum, while the fibrin separates in delicate but dense flakes; the greater density of the small *coagula* renders it difficult, however, to wash away the blood-corpuscles enclosed in these flakes, or to obtain the fibrin as free from hæmatin as that which is obtained from the blood-clot. In order to cleanse the fibrin as much as possible, it is necessary, first to knead it for some time in water, and then to hang it in water in a bag of linen, by which means the salts and the pigment gradually dissolve, and the particles of the fluid rendered thus heavier sink to the bottom of the vessel, while pure water rises in their place.

In order to obtain boiled fibrin in the greatest possible purity, we must dry it after it has been boiled in water; it should be then pulverized and extracted with alcohol containing sulphuric acid, in order to remove any remains of pigment, and finally with ether for the removal of the fat.

*Tests.*—It is only seldom that a case occurs in which any question can arise, as to whether the substance separated from an animal fluid is, or is not fibrin; thus, by way of illustration, the plasma surrounding the organs of insects coagulates on exposure to the air, and we may term this substance fibrin; yet it is by no means identical with the fibrin of vertebrate animals; for it does not separate under the microscope into threads, and it is insoluble in saline solutions, and even in solutions of the alkaline carbonates. Pathological fluids, on exposure to the air, occasionally deposit a sediment. But here the form of the coagulum as well as the microscopic texture of the sediment must decide whether or not the substance which is separated is fibrin; and the action of salts upon it may be observed in the further investigation. In many cases of this kind the separated substance is not fibrin, but consists of albuminous products, which appear under the microscope as minute masses or molecular granules, and whose chemical characters may be recognized by their behavior with hydrochloric and nitric acids and other reagents; or finally, it often consists of fatty or earthy matters that can easily be distinguished by the ordinary tests from true fibrin.

It is often perfectly impossible to distinguish coagulated fibrin from other protein-compounds; and we are therefore not justified in regarding every insoluble mass contained in an exudation as fibrin: the fibrin has, in these cases, already assumed an organized condition, and exhibits the elements of tissues under the microscope; or we find an unorganized, amorphous mass, which is usually not fibrin, although it may be a derivative of that body, and exhibits no property of fibrin that is not common to all the protein-compounds, as we see, for instance, in tubercular

deposits. Many obvious reasons conspire to render a quantitative analysis impracticable in determinations of this kind. It is therefore unchemical, to say the least, for pathological anatomists to designate every unorganized exudation as fibrin; nor shall we learn to distinguish the chemical substrata of these exudations until we shall have thoroughly investigated, in a chemical point of view, the actual constitution of the protein-compounds.

The *quantitative determination of fibrin* in animal fluids has probably been more frequently attempted than that of any other substance; but we nevertheless are still without any method that fulfils the requirements of a good analysis. The usual method of determining fibrin quantitatively is by pressing the clot and washing out the blood, or more frequently by shaking or whipping blood before it has coagulated, and drying and then weighing the fibrin thus separated. In the former case, notwithstanding the most careful washing, the membranous cell-walls and the nuclei (if indeed they exist) of the red blood-corpuscles remain mixed with the coagulum; and there are also technical reasons why this method of treating the blood-clot should occasion a loss of fibrin; hence the second method is generally preferred. We have already seen that fibrin obtained by whipping always contains fragments of some of the red corpuscles and most of the colorless corpuscles; indeed the fibrin thus obtained is far more difficult to wash, and much less compact in its texture, than that which is obtained from the blood-clot; it becomes somewhat reddish on exposure to the air, and often begins to putrefy before it has been freed from all soluble substances. The fibrin determined in quantitative analyses of blood and lymph is never or very rarely free from the fat which adheres most tenaciously to it. Moreover, in some forms of disease, and in certain animals, the blood, when allowed to stand, deposits a flocculent fibrin, which on washing, passes to a greater or less degree through the filter.

If the separated fibrin were always of the same consistence, and if one and the same relation existed in every specimen of blood between the fibrin, the fats, and the colorless corpuscles, we might regard the analyses of different specimens of blood, in reference to the amount of fibrin, as always admitting of comparison; but we know that even under strict physiological relations, the quantity of the lymph-corpuscles suspended in the blood is extremely variable, and thus, for instance, we cannot strictly compare analyses of the blood after repeated venesections (when the blood always contains a very large number of colorless corpuscles) with those of blood not thus modified by venesection.

#### *Physiological Relations.*

*Occurrence.*—The substance which on coagulation forms fibrin occurs principally in the blood, in the lymph, and in the chyle.

Its amount in normal venous blood scarcely reaches 0.3%; according to most observers it fluctuates between 0.19 and 0.28%. In the blood of healthy men Scherer<sup>1</sup> found from 0.203 to 0.263%. This substance has, however, a higher importance than from its small amount we should at

<sup>1</sup> Haeser's Arch. Bd. 10, S. 50.



first suppose, seeing that, in different physiological and pathological conditions, its quantity is liable to greater variations than that of any other constituent of the blood. Even in *different vessels* the blood contains different quantities of fibrin, although the question whether venous or arterial blood contain the greater quantity is still unanswered; at all events the blood of the portal vein contains a far less quantity of fibrin than that of the jugular veins; according to the numerous investigations of Schmid,<sup>1</sup> it is at least three times smaller in the former than in the latter. From some observations of Zimmermann,<sup>2</sup> it appears that the blood in the veins remote from the heart is richer in fibrin than that in the veins nearer to the central organ of circulation. I<sup>3</sup> could not find a trace of fibrin in the blood of the hepatic veins of the horse, while the portal blood was always tolerably rich in that constituent. Funke,<sup>4</sup> in his examination of the blood of the splenic vein, only found a little fibrin in a few cases.

*Sex* appears to induce no difference in reference to the amount of fibrin in the blood, although the quantity of this constituent is affected both by the *period of life* and by *pregnancy*. According to the experiments of Nasse and the more recent investigations of Poggiale,<sup>5</sup> the blood of new-born infants contains less fibrin than that of adults, the augmentation in the amount of fibrin being especially striking at the period of puberty. In pregnancy, as appears from the researches of Andral and Gavarret, it is principally in the last three months that the quantity of fibrin increases. During an *animal diet*, I found that my blood contained a larger amount of fibrin than during a *vegetable diet*; and Nasse<sup>6</sup> has made experiments on dogs with a similar result. There are moreover many corroborative proofs of the correctness of Nasse's observation that the quantity of fibrin in the blood is increased during prolonged *fasting*.

The results independently obtained by Nasse<sup>7</sup> and Poggiale agree in showing that the blood of *herbivorous animals* generally contains more fibrin than that of the *carnivorous* (dogs and cats), and that the blood of *birds* contains even more than that of the *herbivora*.

The results of the quantitative determination of the fibrin in the blood in different forms of *disease* are very numerous, and on the whole tolerably accordant. The most constant and the most decided augmentation occurs in inflammatory diseases, and especially in acute articular rheumatism; in the last-named disease the fibrin has been found to reach 1.18%, and in pneumonia 1.01%.

It is moreover worthy of remark that inflammation in which no fever is present, and likewise mere fevers without inflammation, augment the quantity of fibrin in the blood.

In other diseases, as for instance, in chlorosis, typhus, tuberculosis, Bright's disease, and carcinoma, there seems only to be an augmentation of the fibrin when an inflammatory complication supervenes; in carci-

<sup>1</sup> Heller's Arch. f. Chem. u. Mikrosk. Bd. 4, S. 97-132.

<sup>2</sup> Arch. f. phys. Heilk. Bd. 6, S. 586-600.

<sup>3</sup> Ber. der. Ges. d. Wiss. zu Leipzig. 1850, S. 136.

<sup>4</sup> Dissert. inaug. de sanguine venæ lienalis. Lips. 1850.

<sup>5</sup> Compt. rend. T. 25, p. 198-201.

<sup>6</sup> Handwörterb. d. Physiol. Bd. 1, S. 148.

<sup>7</sup> Journ. f. pr. Ch. Bd. 28, S. 146 ff.

noma, however, certain observations of Popp and Heller appear to indicate that there is a decided augmentation of the fibrin independently of any inflammatory fever.

There are no diseases in which we find a constant and certain diminution of the fibrin; and whenever we find any diminution of the fibrin it is always very slight.

It is however true that in diseases where a constant diminution of the fibrin has been supposed to exist, we have only rare opportunities of analyzing the blood.

In the *lymph* of man Marchand and Colberg<sup>1</sup> found 0.052%, and in that of the horse Geiger and Schlossberger<sup>2</sup> found 0.04% of fibrin.

In the *chyle* of a horse Simon found 0.075%, and in that of a cat Nasse found 0.13% of fibrin.

The fibrin in the *muscles* is by no means perfectly identical with spontaneously coagulated fibrin; it is one of the many species embraced under the generic name of fibrin.<sup>3</sup>

*Syntonin* is the name I have proposed for the substance which Liebig,<sup>4</sup> who was the first to describe it accurately, has termed *muscle-fibrin*. The following are its leading *properties*. When moist, it forms on the filter a coherent, somewhat elastic, snow-white mass, which may be detached from the filter in plates or membranes; by extension and careful teasing, these delicate plates may be made to assume a fibrous appearance under the microscope, not unlike that of the blood-fibrin. The substance, when still moist, dissolves very readily in lime-water as well as in dilute solutions of the alkalis; it coagulates from the solution in lime-water on boiling, in the same manner as albumen; it is precipitated both from this and from the alkaline solutions by concentrated solutions of the neutral salts of potash and soda; the mass swells in a moderately concentrated solution of carbonate of potash, becomes gelatinous and dimly transparent, but does not dissolve; it is only after very considerable dilution that even a portion of the substance undergoes solution. If to the alkaline solutions of this substance we add chloride of calcium or sulphate of magnesia, we obtain no precipitate, unless we boil the mixture; if, however, we have previously boiled the alkaline solution (which at most only induces a slight turbidity), the solutions of the above-mentioned salts then at once induce a flocculent precipitate. Nitric acid throws down a white flocculent precipitate from the alkaline solutions of syntonin; chromic acid, or acid chromate of potash and hydrochloric acid, throws down this substance in flakes both from alkaline and from acid solutions; pure hydrochloric acid, even when added to excess, only renders the alkaline fluid opalescent. I was unable to dissolve uncoagulated syntonin in nitre-water (consisting of 6 parts of  $\text{KO.NO}_3$  to 100 parts of water), even after five days' digestion at 30°.

With regard to its *composition*, Strecker<sup>5</sup> has found in this substance 1.4% of ash (from hens' flesh), 54.46% of carbon (from beef), and 53.67%

<sup>1</sup> Pogg. Ann. Bd. 43, S. 625-628.

<sup>2</sup> Arch. f. phys. Heilk. Bd. 5, S. 892-896.

<sup>3</sup> [Liebig has recently published a memoir on the fibrin of muscular fibre, in which he indicates several points in which it distinctly differs from the fibrin of the blood. See Ann. d. Ch. u. Pharm. Bd. 73, S. 125.—G. E. D.]

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 73, S. 125-129.

<sup>5</sup> Ibid. Bd. 73, S. 127.

of carbon (from mutton), 7.27% of hydrogen, 15.84% of nitrogen (from beef), and 16.26% of nitrogen (from mutton), and from 1.02 to 1.21% of sulphur. This substance is therefore sufficiently distinct in its composition from blood-fibrin. My analyses of the smooth muscles of the stomach of the pig, and of the middle arterial coat of the ox, agree tolerably closely with the analyses of Strecker. Walther<sup>1</sup> found rather more sulphur, namely, 1.6%.

The *preparation* of this substance is best effected in the following manner. We take flesh as free as possible from fat, mince it finely, repeatedly stir it with water, and press it till the fluid which comes off no longer has an acid reaction or becomes turbid on boiling. The mass of flesh, which has been thus washed out, is then stirred with water to which 1-1000th of hydrochloric acid has been added. The fibre-substance of the muscles dissolves very readily in this fluid. On the neutralization of its acid, the filtered fluid at first only yields a turbid jelly, so that the whole fluid either vibrates like freshly solidified glue, or presents a viscid semi-liquid condition; the jelly gradually condenses, and there sink to the bottom white, partially translucent flakes, which must be most carefully washed.

With regard to the *tests* for this substance, we may observe, that notwithstanding its many points of resemblance to albumen and blood-fibrin, it differs from them so essentially in some of its properties, that an error of diagnosis in this direction is hardly probable. Its behavior towards water containing hydrochloric acid (in which blood-fibrin does not dissolve, but only swells), and towards nitre-water and carbonate of potash, will prevent it from being confounded with blood-fibrin; while its precipitability from alkaline solutions by the chlorides of potassium and sodium, or by other salts of the alkalies, sufficiently distinguishes it from ordinary albumen.

The *occurrence* of this body, as the most essential constituent of the fibrillæ of the transversely shaped muscles, was first recognized by Liebig. I have found it not only in the ordinary smooth (unstriped) muscles of the stomach, the intestinal canal, and the urinary bladder, but also in almost all the contractile tissues in which Kölliker has detected the so-called contractile fibre-cells, as for instance, in the middle arterial coat, and in the spleen.

We are unable to form any definite opinion regarding the *origin* of the syntonin from the albuminous matters of the food, or from the albumen or fibrin of the blood, until we possess more distinct knowledge respecting the chemistry of the protein-bodies.

The *uses* of this substance are sufficiently obvious from the parts in which it occurs; it is the main constituent, and the most essential substratum of all the contractile tissues. We are, however, as yet unable to decide as to the extent to which it is more capable of contributing to vital contractility than the other protein-bodies.

We shall return to the consideration of the fibrin of the muscles (muscle-fibrin) when we treat of chemical histology; since, in order correctly to understand its relations, we must have an accurate knowledge of the histological elements of muscular tissue.

<sup>1</sup> Diss. inaug. de musculis laevibus. Lips. 1851.

The remarks which have been already made on the manner of recognizing fibrin, include all that need be stated in reference to the views advanced regarding the coagulated fibrin assumed to be deposited in tissues or exudations.

In the preceding description of fibrin, a criticism might probably have been expected on the several varieties of this substance, which have been described by different writers as occurring in morbid fluids; we have, however, made no reference to Nasse's fibrin-disks, to Zimmermann's molecular fibrin, to Rokitsansky's pseudo-fibrin, or to the fibrin of later coagulation, to which Virchow attaches much importance, because we regard discussions on such points as out of place in the department of strict zoo-chemistry; for it is only after the principles of zoo-chemistry have been fully discussed, and when we enter on the theory of the animal juices, that we can form a sound judgment on such subjects.

*Origin.*—Taking into consideration everything connected with the occurrence of fibrin, we can scarcely entertain a doubt that it is formed from albumen, and not directly from the protein-containing food; for its occurrence in the chyle is not opposed to this view, partly because, as Henle has shown, fibrin may be conveyed to this fluid by the lymphatics and bloodvessels, and partly because, as I have fully convinced myself, all the juices of the animal body not only contain free carbonic acid but also free oxygen. It was formerly supposed that the formation of fibrin from albumen might very easily be accounted for; since, according to the older analyses of Mulder, fibrin contained one-half less sulphur than the albumen of serum, nothing seemed more simple than to assume that the oxygen conveyed by the respiration to the blood, converted half of the sulphur of the albumen into sulphuric acid, and that this combined with its alkali, so that fibrin was now evolved. These and all similar views have become untenable since more recent analyses of albumen and fibrin have been made. If we would at present start an hypothesis regarding the formation of fibrin, it can only rest on the slight excess of oxygen which fibrin contains over albumen. The indication afforded by this fact has led, however, to serious error in reference to the increase of the fibrin in inflammations: since it was concluded that, although we may not know how the oxygen finds its way to the albumen to form fibrin, it is at all events incontestable that the latter is formed by a process of oxidation or cremacausis; and it was further very erroneously concluded that the augmentation of the fibrin in inflammation is dependent on an increased rapidity of the process of oxidation, and that consequently inflammation is nothing more than an actual process of combustion. This hypothesis, originally propounded by chemists, was for a long period accepted by physicians, without any doubts occurring as to its correctness. In accordance with chemical principles, an excessive supply and absorption of oxygen *might* indeed be regarded as the cause of an increase of fibrin; but even this is by no means proved; for how would it then be possible that in pneumonia, where a greater or lesser part of the lungs is hepatized, that is to say, is rendered impermeable to air, a greater quantity of fibrin should be found in the blood than during other inflammatory affections? This has lately been referred to the greater frequency of the respirations, but independently of the circum-

stance, that in inflammation of other parts, the number of the fibrin should then attain at least the same height as in pneumonia, we know that fever, notwithstanding it is often accompanied by an increased frequency in the respirations, by no means gives rise to an augmentation of the fibrin. Physiological facts lead us to exactly the opposite hypothesis, that *the augmentation of the fibrin in inflammatory blood is to be referred to a diminution in the supply of oxygen*. The frequent but short and incomplete respirations which occur only in febrile (and not in non-febrile) inflammations, are only sufficient to convey to the blood sufficient oxygen to convert certain substances into fibrin but not to oxidize them further; this is the reason why the amount of fibrin attains its *maximum* in pneumonia and pleuritis, and why the blood in the former disease is most rich in carbonic acid, for this gas is scantily excreted in proportion as oxygen is scantily received by the lungs. The physiological importance of fibrin affords arguments altogether in favor of this view.

*Uses.*—The phrases, *progressive* and *regressive* metamorphosis, of whose import we have spoken in an early part of this volume (see p. 37), have led to a long contest regarding the physiological importance of fibrin. On the one hand, it has been correctly maintained that this substance must be necessary to the formation of tissues, since, as a general rule, the only exudations which are capable of organization are those which contain fibrin; on the other hand, stress is laid upon the circumstance that an augmentation of the fibrin coincides with those states in which nutrition and renovation are most affected, and on the incontestable fact that the fibrin in the blood is found to be increased when more albuminous matters have been taken as food than could be applied to the reparation of effete tissue. We regard it, however, as superfluous to enter into the detailed arguments for and against these two opinions. The bearing of the whole case is simply this. It is pretty well established that fibrin is formed by a process of oxidation from albuminous matters; now we know that almost all the tissues are richer in oxygen than fibrin, and on the other hand, that the effete materials of tissue and the excess of nutrient matter can only be removed from the system, that is to say, be converted into ordinary excreta, by oxidation. Hence, the simplest view is to regard fibrin as representing a transition stage. If an albuminous body in the animal organism be more highly oxidized, it cannot altogether exceed the transition stage which is represented by fibrin, although indeed, the formation and increase of the latter may not always be evident. An analogous instance from pure chemistry will elucidate this view; we know, from Liebig's celebrated investigations on fermentation, the intermediate stages which during the process of acid fermentation present themselves between the two extremes of spirit of wine and acetic acid. We know that by a gradual process of oxidation, aldehyde and aldehydic acid are formed from the spirit, although these two substances may not become apparent: the beautiful investigation of Mulder regarding the *Mycoderma aceti* affords an almost more analogous illustration; its cellulose can only be produced by a process of oxidation from the alcohol; moreover, in the formation of this cellulose from the alcohol there must first be formed

an aldehyde-like substance poorer in oxygen than cellulose; hence aldehyde may just as well be produced during the oxidation of alcohol into acetic acid, as during its oxidation into cellulose. In a perfectly analogous manner we may regard the fibrin as representing one of the stages in the oxidation of the albumen, which is transferred either into the tissues or into the secreted substances. There seems to us to be no discrepancy between the above observations on the chemical importance of fibrin, if we will only leave nature unfettered with divisions into progressive and regressive metamorphoses. For, if we assume the formation of tissue to be the highest stage of animal metamorphosis, fibrin pertains to the ascending or progressive series, inasmuch as it yields the proximate stratum for the development of cells and the formation of tissues; on the other hand, it must be classed in the descending or regressive series, in so far as its quantity in the blood is found to be increased in diseases, or after the excessive use of albuminous food, when it does not become converted into tissue but is changed by oxidation into the ordinary excreted matters. For we cannot believe that, as in the percussion-apparatus of Physicists, a given quantity of fibrin will repel and displace a corresponding amount of tissue. In short, we seem to be nearest the truth in regarding fibrin as representing one of the most common stages in the metamorphosis of albuminous substances.

We must not conclude our observations on fibrin without noticing a very common error that has crept into pharmacology from the misunderstanding of a chemical fact. Many physicians believe that the antiphlogistic power of nitrate of potash is explained by the chemical fact that spontaneously coagulated fibrin dissolves in a solution of nitre. Without entering into the question whether this salt actually possesses the power ascribed to it, we assert that this mode of explanation is altogether untenable, for it is difficult to draw the conclusion that nitre can prevent the formation or augmentation of fibrin in inflammatory blood, simply because coagulated fibrin is soluble in a solution of this salt. According to Scherer, the fibrin of inflammatory blood appears to be insoluble in this saline solution; how then can a solution of nitre prevent the augmentation of fibrin in inflammatory blood through a solvent power which, in relation to this inflammatory fibrin, it actually does not possess?

There would be much more probability in the assumption that a solution of nitre hindered the coagulation of highly fibrinous blood, or that it redissolved already coagulated fibrin. The most simple arithmetical example will illustrate this view. Scherer asserts that 1 part of nitre is required to dissolve 1.5 parts of fibrin; assuming that the quantity of the blood amounts to twenty pounds, and that it contains only 0.3% of fibrin, the whole amount of fibrin would be not less than 300 grains, and to dissolve this quantity 200 grains of nitre should be at once taken; physicians, however, usually prescribe about 10 grains every two hours, so that in 24 hours 100 or 120 grains are at most all that is taken to act upon the fibrin. But the amount of nitre in the blood can never rise even to this insufficient height, partly because the salt becomes distributed from the bloodvessels into the juices of the body generally; and partly because it is much too rapidly carried off by the urine to admit

of its accumulating in great quantity in the blood. Even if it were possible to prove that nitre possesses this power, it would be very singular and inexplicable why we never class amongst the special antiphlogistic medicines other salts, as, for instance, the alkaline carbonates, which possess a much greater power of dissolving fibrin, and of preventing its coagulation.

In this pharmacological digression, we cannot help remarking that if inflammation were actually a process of oxidation or combustion, it is very strange that we have not found the alkaline salts of the vegetable acids, the *amylacea*, and the fats, to be the most efficient antiphlogistics. It is true that we attack severe inflammation with tartar emetic, but even when given according to Rasori's method, it communicates to the blood so little combustible material as to be inappreciable, especially when combined with an antiphlogistic diet. If inflammation were a process of combustion, the antiphlogistic diet must be exactly the reverse of that which we understand by the term. Moreover, direct experiments on patients, to whom large doses of acetate and tartrate of potash might safely be administered, have proved that these salts exert no action either of a beneficial or of an injurious character, on the inflammatory process. Even the most zealous adherents of the chemico-pathological theory of combustion would hardly attempt to regard the fat in the emulsion as an antiphlogistic, since it has been already proved by Nasse and others that the fibrin of inflammatory blood, and of the *crusta inflammatoria*, contains nearly twice as much fat as ordinary fibrin, unless, indeed, he would attempt to trace to this fact the *digitus index medicatricis naturæ*, protecting the fibrin from the action of the oxygen through the agency of combustible fat.

## VITELLIN.

### *Chemical Relations.*

*Properties.*—This is the albuminous body of the yolk of egg; it is so similar to albumen that, until recently, it has been confounded with the albumen of the white of egg; like the latter, it exists both in a *soluble* and in an *insoluble modification*; the former is not precipitated from its aqueous solution by organic acids or by ordinary phosphoric acid, but is thrown down by sulphuric and hydrochloric acids; at 60° its solution begins to become opalescent, and at from 73° to 76° there is a deposition of larger or smaller flakes. It is only distinguished from soluble albumen by the circumstances that (without the addition of acetic acid or of salts) when heated, it forms flakes and clots, that it is not precipitated by the salts of oxide of lead or of copper, and that it is thrown down by ether.

*Coagulated vitellin* has the same properties as coagulated albumen, and the similar modifications of the other protein-compounds. Moreover, in its reactions it coincides with Mulder's binoxide of protein or fibrin-protein.

*Composition.*—Dumas was the first who analyzed this body, and discovered that it differed from albumen; according to this analysis, with

which that subsequently made by Gobley<sup>1</sup> very well agrees, vitellin contains 3 atoms of water more than albumen; according to Gobley it also contains phosphorus and sulphur. Mulder, and especially v. Baumhauer,<sup>2</sup> have subsequently made accurate analyses of this body, and regard it as a combination of oxide of protein with sulphamide, so that its theoretical formula would somewhat resemble that of fibrin. According to v. Baumhauer, the phosphorus contained in vitellin exists in it solely in the form of phosphate of lime; moreover his amount of sulphur is obviously too small, since he only determines this substance in the moist way.

To give a general idea of the composition<sup>3</sup> of this body, we append the mean numbers obtained by Gobley and by v. Baumhauer.

	Gobley.	v. Baumhauer.
Carbon, . . . . .	52.264	52.72
Hydrogen, . . . . .	7.249	7.09
Nitrogen, . . . . .	15.061	15.47
Sulphur, . . . . .	1.170	0.42
Phosphorus, . . . . .	1.020	—
Oxygen, . . . . .	23.236	24.30
	100.000	100.00

Berzelius conjectures that we are here not dealing with a simple substance, but with an admixture of substances, as is unfortunately the case with most of the protein-compounds. Vitellin, extracted with indifferent menstrua, contains 4.043% of phosphate of lime.

*Preparation.*—Soluble vitellin, in a pure state, that is to say, free from yolk-fat and from yolk-globules, has not yet been exhibited. Gobley has only attempted to ascertain its reactions after stirring the yolk of egg with water and allowing the emulsive constituents, as much as possible, to deposit themselves. In its *coagulated* form we can obtain it in a far purer state; boiled and triturated yolk of egg is extracted with ether, alcohol, and water, then dissolved in acetic acid, and precipitated therefrom by ammonia, with, however, such precaution that the fluid remains sufficiently acid to retain the phosphate of lime in solution; the gelatinous precipitate is then dried and extracted with water and alcohol.

*Tests.*—The methods of recognizing and quantitatively determining vitellin are sufficiently obvious from our description of the properties of this body.

#### *Physiological Relations.*

*Occurrence.*—Hitherto vitellin has only been recognized in the yolk of egg, of which, according to Berzelius,<sup>4</sup> it constitutes about 17%, or, according to the most recent investigations of Gobley, 15.76%. No eggs but those of the common hen have as yet been examined.<sup>5</sup>

<sup>1</sup> Journ. de Pharm. T. 11, pp. 410–17, et T. 12, pp. 5–12.

<sup>2</sup> Scheik. Onderzoek. D. 3, p. 272, or Arch. der Pharm. Bd. 45, S. 193–220, and Unters. H. 2, S. 80.

<sup>3</sup> [Vitellin has also been recently analyzed by Noad. See the Chemical Gazette, vol. 5, p. 409.—G. E. D.]

<sup>4</sup> Jahrb. d. Ch. Bd. 9, S. 650.

<sup>5</sup> [Gobley has recently examined the eggs of the carp, which in their chemical composition seem very similar to those of the common hen. Journ. de Chim. Méd. T. 6, p. 67.—G. E. D.]



*Origin.*—It is very easy to conceive that vitellin may be formed from albumen or fibrin, but in the yet imperfect state of our knowledge regarding albumen and fibrin as well as vitellin, we cannot chemically trace out this metamorphosis. Since, however, it is poorer in carbon, and somewhat richer in oxygen, than albumen, it may, like fibrin, be regarded as one of the first stages of the metamorphosis of albumen by the action of oxygen, and as a certain form of non-spontaneously coagulating fibrin.

*Uses.*—From the position in which vitellin occurs and from its analogy with other albuminous substances, it is obviously one of those nutrient substances which are employed in the formation of the animal tissues. We are however entirely ignorant of the chemical equations representing these changes; from the admirable work of Baudrimont and Martin St. Ange<sup>1</sup> we may however at least draw the conclusion that this substance loses a portion of its nitrogen and assimilates oxygen in its conversion into tissue. (See the “History of Development,” in the second volume.)

## GLOBULIN.

### *Chemical Relations.*

*Properties.*—This body, which has also received the name of *crystallin*, occurs naturally in the soluble state, but becomes insoluble on boiling. Soluble globulin, when dried at 50°, forms a yellowish, transparent mass, which may be easily triturated, and then yields a snow-white powder; it is devoid of smell and taste, swells like albumen in water, and gradually dissolves, forming a viscid solution containing merely a few flakes; after precipitation by alcohol from this solution, it is insoluble in water, but, like casein, is partially soluble in boiling alcohol; on cooling, however, it again separates from this solution. The aqueous solution of globulin is coagulated by ether. When dried, the soluble modification may be heated to 100° without passing into the insoluble state. It is distinguished from albumen and vitellin, which are very similar to it, by the following properties: its solution does not become opalescent at a lower temperature than 73°; at 83° it assumes a milky turbidity, and at 93° separates as a globular mass (if it be still mixed with hæmatin) or as a milky coagulum which never becomes clear on filtration, and from which neither small quantities of acetic acid or ammonia separate flakes capable of being removed by filtration; it is only when neutral alkaline salts are added, and the solution is then boiled, that the fluid becomes perfectly clear and flakes and small clots are deposited. The following reaction is very characteristic of globulin: its solution is not precipitated either by acetic acid or by ammonia, but it becomes strongly turbid when the fluid treated with acetic acid is neutralized with ammonia, or conversely when after the addition of ammonia it is neutralized with acetic acid. Its behavior simply with acetic acid is, however, also different from that of albumen. On the addition of a little dilute acetic

<sup>1</sup> Ann. de Chim. et de Phys. T. 21, pp. 195–257.

acid, the solution of globulin becomes opalescent, and when heated to  $50^{\circ}$  a milky coagulum separates; the fluid rendered turbid by a little acetic acid, becomes clearer when more of the acid is added, but always remains opalescent; this fluid does not coagulate till heated to  $98^{\circ}$ ; it is only when a very great excess of acetic acid has been added that the globulin ceases to be coagulable by heat. The behavior of globulin towards mineral acids and metallic salts is precisely the same as that of albumen. It is also coagulated by creosote; it decomposes and becomes putrid much more readily than the other protein-compounds; when boiled it develops ammonia.

Lecanu regarded this body as identical with albumen, Simon with casein; we would rather place it by the side of vitellin, if the elementary analyses were not opposed to this view; but it appears to us by no means advantageous to science, to group together several ill-defined substances merely on the strength of a few reactions, and without any definite proof of their similarity.

Berzelius ascribes\* to the globulin, united in the blood with hæmatin, the singular property of dissolving in water containing albumen and little or no salts, but not in water which holds in solution large quantities of alkaline salts. He was in error in regarding the sediment of the blood-corpuscles, which he named hæmato-globulin, as a simple mixture of globulin and hæmatin; for we shall show (in the section on "The Blood"), that this hæmato-globulin is composed of blood-corpuscles which by the law of endosmosis become so distended in pure water as scarcely to be visible under the microscope, but which (unless the blood-corpuscles have burst from too great an addition of water) again become apparent when we add a salt to the fluid in which they are immersed, and thus render it denser; in which case the blood-corpuscles again contract, become denser and flatter, and are again visible.

No properties have yet been detected in coagulated globulin by which it may be distinguished from other boiled protein-compounds.

*Composition.*—Globulin has been subjected to even fewer analyses than vitellin; as that which is contained in the blood can never be perfectly freed from hæmatin, no accurate analysis can be made of it. Dumas<sup>1</sup> has however analyzed a specimen containing hæmatin, while both Mulder<sup>2</sup> and Rüling have analyzed this substance as obtained from the crystalline lens.

	Mulder.	Rüling.
Carbon, . . . . .	54.5	54.2
Hydrogen, . . . . .	6.9	7.1
Nitrogen, . . . . .	16.5	} 37.5
Oxygen, } . . . . .	22.1	
Sulphur, } . . . . .		1.2
	<hr/> 100.0	<hr/> 100.0

Although Berzelius assumed that phosphorus as well as sulphur was contained in this substance, Mulder found only the latter, which averaged

<sup>1</sup> Compt. rend. T. 22, p. 904.

<sup>2</sup> Journ. f. pr. Ch. Bd. 19, S. 189; and Bullet. d. Néerl. 1839, p. 196.

0.265% : this sulphur was however determined in the moist way; in the dry way, I determine the sulphur in globulin from the crystalline lens of the calf (as a mean of three experiments) at 1.134%, and Rüling<sup>1</sup> in globulin similarly obtained from the ox at 1.227%. Mulder, at present, regards globulin as a combination of his hypothetical protein with sulphamide.

The globulin of the crystalline lens contains only a very small amount of insoluble ash-constituents; I found only 0.241% of phosphate of lime.

In globulin from the crystalline lens of a calf I found 1.548% of soluble salts consisting of metallic chlorides, sulphate of soda (= 30.37% of the soluble salts) and alkaline phosphates (= 7.77% of the soluble salts), but containing no alkaline carbonates. On the other hand, on evaporating the fluid filtered from the coagulated globulin (which besides 92.095% of coagulated globulin yielded 7.905% of soluble residue) I obtained on the incineration of this residue an ash which contained only 13.166% of phosphate of lime, while the soluble salts contained a large quantity of alkaline carbonates, namely 16.71%.

Now as the ash of non-coagulated globulin contains no alkaline carbonate, we may conclude that in soluble globulin soda is combined with an organic substance—either with the globulin itself or with an organic acid,—and that after the destruction of the globulin, this free alkali combines with the sulphuric acid produced from the globulin, which would account for the circumstance, that the ash of the collective globulin contains no alkaline carbonate; if, on the other hand, the soluble salts are separated from the globulin on its coagulation (in the same manner as albumen on coagulation loses its alkali) they contain much alkaline carbonate after the combustion of the organic substance not separated with the coagulated globulin, for here there is no formation of sulphuric acid to decompose the alkaline carbonates. No alkali occurring in the ash as a carbonate, can, according to my view, be combined with the globulin previously to its coagulation, for the following reason. The solution of globulin from the crystalline lens has a distinct, although a very faint *alkaline* reaction; during the process of coagulation, we may easily show that it develops ammonia, and afterwards the fluid does not, as in the case of albumen, exhibit a stronger alkaline reaction, but on the other hand is now *acid*; this phenomenon cannot be more simply explained than by the assumption that there is *phosphate of soda and ammonia* in the fluid, for the solution of this salt has an alkaline reaction, loses ammonia on boiling, and finally assumes an acid reaction when the salt is thus converted into acid phosphate of soda. Now, if globulin were contained in this fluid, no acid reaction could ensue after its coagulation, because the soda separated from the globulin would take the place of the ammonia that escaped from the phosphate. Hence this soda which is combined with carbonic acid in the ash of the residue from which all globulin has been removed, must have been previously in combination with an organic acid. If, for the present, we regard this organic acid as lactic acid, until the subject can be more

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 313.

accurately investigated, we can scarcely be charged with adopting too bold an hypothesis, since this acid cannot at all events be one of the volatile acids of the animal body. We are, unfortunately, still compelled to rest upon such deductions as these, in our endeavor to investigate the nature of the salts held in solution in association with animal substances, since, as we shall subsequently see (when treating of "the mineral constituents of the animal body,") the constituents of the ash unfortunately afford very little information regarding the actual constitution of the salts that existed previously to the calcination of the residue. I must, moreover, remark that the boiling must be continued for some time, in order that the acid reaction after the coagulation of the globulin may manifest itself.

*Preparation.*—As in the case of soluble albumen, it is impossible to prepare *soluble globulin* in a perfectly pure state. Globulin presenting the reactions which we have already indicated, may be obtained by neutralizing with acetic acid the fluid of the crystalline lens, evaporating it to dryness at a temperature not exceeding 50°, and extracting the residue with ether and dilute alcohol. The globulin of the blood, which cannot be separated without decomposition of the hæmatin, presents, with the exception of its color, exactly the same relations as the globulin obtained in the above manner from the crystalline lens.

Mulder prepared *coagulated globulin* by simply extracting with alcohol and ether, globulin which had been precipitated by boiling. The coagulated globulin which I examined was precipitated with hydrochloric acid, washed with the same acid, then dissolved in water, again precipitated by carbonate of ammonia, and finally washed with water, alcohol, and ether, after which it left no perceptible ash.

*Tests.*—In the preceding remarks we have mentioned the reactions by which globulin may be distinguished from the similar protein-compounds: we will here merely add that no other *soluble* protein-compound is precipitated both from its acid and its alkaline solution by neutralization, although almost all the insoluble protein-compounds possess this property—a circumstance which affords a proof that globulin is reduced to the coagulated state, both by an excess of alkali and by an excess of acid. In our observations on casein, we shall point out how it may always be distinguished from that substance. It will always be difficult—indeed at present it is impossible—to recognize globulin with certainty when it is mixed with albumen or casein. Here, unfortunately, elementary analysis affords us no assistance, since it so closely approximates in its ultimate constitution to other protein-compounds.

In attempting a *quantitative determination* of globulin we must adopt the same precautionary measures as in the determination of albumen; indeed, as we have already shown, there are even greater difficulties in reducing globulin to a condition in which it can be easily and thoroughly collected on a filter, than are presented by albumen. We must acidify with acetic acid and apply heat; then saturate the acid with ammonia, and boil strongly and for a considerable time, in order to obtain the globulin in a state admitting of its being readily collected on a filter. Even if we succeeded in distinguishing globulin from any similar body, as, for instance, albumen, by its relation to acetic acid, and by noticing

its behavior when heated to  $50^{\circ}$  (see p. 330), or by observing that it was precipitated by the neutralization either of its acid or its alkaline solution, we could not by these means separate it from that body; for it would not be in a state fit for filtration, that is to say, it would either pass through the filter in a turbid condition, or it would stop up the pores of the filter, and could not by any possibility be washed off.

### *Physiological Relations.*

*Occurrence.*—Globulin occurs in the cells of the *crystalline lens*, in a very concentrated solution. In the human lens Berzelius<sup>1</sup> found 35.9% of dry globulin.

Globulin is one of the principal constituents of the *blood*, since, with hæmatin, it forms the viscid fluid contents of the blood-corpuscles.

We can form no definite and certain idea, regarding the quantity of globulin contained in the blood-corpuscles, for even if we are able to form an approximative idea of the amount of hæmatin contained in the corpuscles (see p. 272), we have no means of deciding how much of the remainder of them (amounting to 94.28%) is to be ascribed to fat, to the enveloping membrane, and to globulin. Hence it is not possible to make any accurate statement regarding the quantity of globulin contained in the blood generally. We shall, however, return to this subject in the second volume, when treating of "the blood-corpuscles."

Globulin has not yet been found in any other parts of the animal body. In the present state of organico-analytical chemistry, we are unable to attempt to seek it in its coagulated state.

*Origin.*—In regard to the seat of the formation of globulin, no reasonable doubt can be entertained, that it at present has only been found in cells and cell-like bodies like the blood-corpuscles.

Whichever view we adopt regarding the mechanical mode of formation of the red from the colorless corpuscles (see p. 273) and the remarks "on the blood-corpuscles," in the second volume, we must arrive at the conclusion, that the globuloid is formed within a cell or a vesicle or a closed saccule, which is bathed in an albuminous fluid. If albumen lies without the enveloping membrane and globulin exists within it, we are almost compelled to assume that the globulin is produced by the cellular action from the albumen, but we cannot give the chemical equation, representing how this transformation takes place, for the simple reason that we are ignorant of the rational composition both of albumen and globulin. From a comparison of the analyses of albumen and globulin, we can, however, perceive that the latter contains a little less carbon and sulphur, but rather more oxygen than the former. (Little weight can be attached to the amount of phosphorus in albumen, in consequence of the uncertainty connected with our modes of determining that element.) Hence globulin appears to be albumen modified by oxidation, so that it is allied to fibrin, or perhaps more correctly should be placed between this substance and albumen. Moreover, the physiological hypothesis, according to which the blood-corpuscles are to be regarded as nothing more than laboratories in which the ordinary nutrient matter,

<sup>1</sup> Lehrb. d. Ch. Bd. 9, S. 528.

crude albumen, is first prepared, in order to become applicable to the formation or reparation of tissues in different organs, corresponds with this view. Whether globulin be directly converted into fibrin, is a question which, at present, is unanswerable; we shall, however, return to this subject in a future part of this work.

*Uses.*—The object of nature in depositing globulin in the cellular fibres of the crystalline lens is too obvious to require comment. It is, however, interesting to observe that nature, in producing a refractive fluid, aimed at rendering the lens achromatic, not merely by anatomical structure, but also by filling its middle layers with a concentrated fluid which is always attenuated toward the capsule.

Chenevix is the first to whom we are indebted for this observation; he found that the specific gravity of a lens weighing 30 grains, taken from the eye of the ox, was 1.0765, while, when he had peeled off the outer layers, the nucleus, weighing 6 grains, had a specific gravity of 1.194.

But how nature, to carry out this object, effects the separation or secretion of pure globulin, free from albumen and hæmatin, in the crystalline lens, from the minute capsular artery, will probably never be understood.

From the above observations, it is manifest that we can never understand the importance and the uses of the globulin in the blood, until we have obtained an accurate knowledge both of its chemical constitution, and of the function of the blood-corpuscles.

## CASEIN.

### *Chemical Relations.*

*Properties.*—In its dry state *soluble casein* occurs as an amber-yellow mass, devoid of odor, insipid and viscous when tasted, and having neither an acid nor an alkaline reaction; it dissolves in *water*, forming a yellowish viscid fluid, which on evaporation becomes covered with a white film of insoluble casein which may be readily drawn off. If a concentrated solution of casein be exposed for a long time to the air, it rapidly passes into a state of *putrefaction*, developing a very large quantity of ammonia, and yielding leucine, tyrosine, and similar substances.

*Alcohol* renders casein opaque, and gives it the appearance of coagulated albumen; a part, however, of the casein dissolves in alcohol, and on evaporation can be again obtained in an unchanged state; in boiling alcohol it dissolves more freely, but on cooling, the greater part of the casein again separates; this casein thus treated with alcohol dissolves tolerably readily in water, especially with the aid of heat, and has all the properties of non-coagulated casein. If we add a little alcohol to a concentrated aqueous solution of casein, a precipitate is thrown down, which, however, dissolves again readily in water; if, however, the precipitation be effected by the free addition of strong alcohol, the casein is then difficult of solution or even insoluble in water. By *boiling* it is not coagulated from its solutions.

*Acids* precipitate casein from its aqueous solution, and partially com-

bine with it, but they do not reduce it to the coagulated state, for on neutralization with alkalis or metallic oxides, the casein again dissolves; these combinations of casein with acids are readily soluble both in pure water and in alcohol. Casein is especially distinguished from albumen by the circumstances that it is precipitated from its aqueous solutions by *acetic* and *lactic acids*, the precipitate not being an acetate or a lactate, but pure casein. The precipitate is only slightly soluble in an excess of acetic acid; like all the other combinations of this class with acids, it is precipitated by ferrocyanide of potassium. The alcoholic solution of casein is not only not precipitated by acids, but alcohol even possesses the property of dissolving those combinations of casein with acids, which are insoluble in water. When treated with concentrated *nitric*, *hydrochloric*, or *sulphuric acid*, casein yields the same products of decomposition as albumen and fibrin. *Tannic acid* precipitates it from very dilute aqueous and alcoholic solutions.

Casein combines very readily with *bases*, turbid solutions of this substance becoming clear on the addition of caustic *alkalies*; *alkaline earths* dissolve in solutions of casein, and can only with difficulty be separated from that body; with larger quantities of these earths casein forms insoluble compounds. Hence its solutions are precipitated by chloride of calcium and sulphate of lime, as well as by sulphate of magnesia, *on the application of heat*, which thus afford a reaction very characteristic of casein. It resembles albumen in being precipitated by *metallic salts*, and forming with them two combinations, namely, one of casein and the acid, and the other of casein and the metallic oxide. Ferrocyanide of potassium does not throw down casein from alkaline solutions, and only induces a slight turbidity in neutral solutions.

These are the properties of casein, as it occurs in its ordinary state of solution in the milk; if, however, we obtain it perfectly free from alkali, according to Rochleder's<sup>1</sup> method, which we shall presently give, it presents some characters different from those which we have just described. For instance, it dissolves only very slightly in pure water, rather better in hot water, and not at all in alcohol; it reddens blue litmus, without, however, communicating this property to water, but it forms solutions with carbonate and phosphate of soda, which no longer exhibit an alkaline reaction; it dissolves very readily in solutions of hydrochlorate of ammonia, nitrate of potash, and other neutral alkaline salts, does not coagulate on boiling, like albumen, but forms on evaporation a film of casein, as we have already described. It dissolves in dilute mineral acids, but is precipitated on the addition of an excess of the acid; the solutions of casein in dilute acids become covered on evaporation with this colorless, transparent, and somewhat tough membrane; the solution of this substance in acids or in alkalis is completely precipitated by neutralization, and mineral acids throw it down from its acetic acid solution. The precipitated hydrochlorate of casein is, like the hydrochlorate of albumen, soluble in pure water; before dissolving, however, it swells, like the latter, into a jelly-like mass; both acids and alkalis precipitate it from this solution; the deposit thrown down by hydrochloric acid swells and finally dissolves in alcohol, but is precipitable from this fluid by

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 45, S. 253.

ether, this precipitate being again soluble in water. The mere boiling of a solution of casein, under no circumstances, induces a precipitation. On the other hand, we may be readily led to believe that it is converted into a coagulable substance when we have dissolved it in a solution of carbonate of potash, or of nitre to which a little potash has been added; on neutralizing this solution with an acid, a transitory precipitate ensues on stirring or shaking the mixture, and if we now boil the fluid, there is formed an abundant thick coagulum; I have not been able to persuade myself to regard this as a modification of casein coagulable by mere heat (such as sometimes appears to be contained in the milk) but I rather incline to the belief that the acid has converted only a part of the caseate of soda occurring in solution, and of the simple carbonate of soda, into acid salts, and that on the application of heat it is only the acid salts remaining in solution which are decomposed and evolve carbonic acid, while the casein is precipitated.

From the above observations it follows that casein is not reduced to its *coagulated* state by the same means as albumen and globulin. We have long been acquainted with the fact that the casein in milk is coagulated by the mucous membrane of the stomach of the calf; our knowledge is, however, by no means clear regarding the peculiar condition under which this coagulation ensues. We have seen that soluble casein, on the evaporation of its solution, is partially transformed into the insoluble modification; cases, however, occur, in which the whole of the casein in milk is rendered insoluble by evaporation. Even on prolonged exposure to the air, it is well known that milk coagulates; the casein thus separated reacts in the same manner as the precipitate obtained from a solution of pure casein by means of lactic acid, that is to say, after treating it with carbonate of lime or baryta, it is only slightly soluble in water, most of it having been transformed into the insoluble modification. Simon<sup>1</sup> and Liebig explain the coagulation of casein by the calf's stomach (rennet) by assuming that the latter primarily acts as a ferment, converting the sugar in the milk into lactic acid, which precipitates the casein; Simon moreover maintains that he has observed that solutions of casein free from milk-sugar are not coagulated by rennet. Certain experiments, instituted by Selmi,<sup>2</sup> are, however, opposed to this view; he found that alkaline milk could be coagulated by rennet in the course of ten minutes, and that, after the coagulation, it still had a decidedly alkaline reaction; the same was observed when milk, artificially rendered alkaline by the addition of soda, was exposed to the action of rennet. Conversely, casein dissolved in an excess of acetic or oxalic acid, coagulated, like the alkaline solution, at a temperature of from 50° to 56°. The true cause of coagulation is still entirely unknown. It appears, however, from the observations of Scherer,<sup>3</sup> that casein cannot coagulate in the form of a membrane, unless in the presence of oxygen.

From the large number of individual facts which we have mentioned in relation to casein, it may be inferred that our knowledge of this substance is still very defective; for otherwise we could have embraced in a

<sup>1</sup> Frauenmilch. S. 29.

<sup>2</sup> Journ. de Pharm. T. 9, pp. 265-267.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 36.



few paragraphs the most essential points in relation to this body: our difficulties are increased by the probability that casein is not to be regarded as a simple organic body, but as a mixture of at least two different substances. Mulder<sup>1</sup> and Schlossberger<sup>2</sup> have especially directed attention to this circumstance. If freshly washed casein be digested for a couple of days with dilute hydrochloric acid, it is found to be perfectly dissolved; by neutralization with carbonate of ammonia there is precipitated from this fluid a white, viscid body, difficult to separate by filtration; but in the neutralized fluid there still remains in solution another substance which may be thrown down by an excess of hydrochloric acid; and the hydrochloric acid even now holds in solution a protein-like body. The first of these bodies was found by Schlossberger to contain sulphur, and the second to be free from that element.

Here, however, it might be supposed that the prolonged digestion of the original casein with the dilute hydrochloric acid had decomposed it into several substances. Another and an earlier experiment of Mulder, however, supports the view that casein consists of several substances. To milk which had been as thoroughly as possible freed from butter-globules by chloride of sodium, Mulder added dilute hydrochloric acid, which yielded the ordinary precipitate; there remained, however, in solution, a similar body, which was not precipitated till this mixture was boiled.

It is very difficult to arrive at a definite opinion on this point; for any one repeating the experiments on casein which have been described by different authors, will find that all the statements regarding this substance confirm one another to a certain degree, but that on often repeating the same experiment differences present themselves which thus explain the discrepancies in the statements of different observers. Casein appears to us to be a highly transmutable substance, often undergoing change on the application of the mildest reagents. In a word, a method of preparing casein, which would exclude all suspicion of its being changed by the process, is still a desideratum. The circumstance that the elementary analyses of the separated matters give such slightly different results, adds very much to our difficulty of ascertaining whether the constitution of casein is simple or complicated.

Casein, when thoroughly *coagulated* by rennet, and purified, is hard, and presents a yellowish translucent appearance; it softens and swells in water, but is insoluble both in that fluid and in alcohol. Like its soluble modification it combines with *acids* and *alkalies*; but on separating the inorganic part from the casein, the latter is insoluble in water. In its relation to the stronger mineral acids it in every respect resembles coagulated albumen; it is as difficult of solution in acetic acid as its soluble modification; alkalies dissolve it very readily, and, if concentrated, decompose it like the other protein-compounds on the application of heat. On *heating* casein, it softens, may be drawn out in threads, and becomes elastic; and at a higher temperature it fuses, swells up, carbonizes, and develops the same products of distillation as albumen and fibrin; when strongly heated in the air it burns with a flame, and,

<sup>1</sup> Berzelius. Jahreshr. Bd. 26, S. 910.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 92-95.

unless carefully washed with acidulated water, leaves an ash containing carbonate and phosphate of lime, but no alkali.

The investigations of Iljenko<sup>1</sup> show that casein during its putrefaction (even when perfectly freed from fat) develops at first carbonate of ammonia and hydrosulphate of ammonia, but that, after a space of from two to five months, its principal products are ammonia, valerianic acid, butyric acid, and leucine, and to these substances Bopp<sup>2</sup> adds a white, crystallizable, sublimable body, having a very strong fæcal odor, and an acid which, when decomposed with a mineral acid, yields a brown substance together with tyrosine, and ammonia. On fusing casein with hydrated potash, it develops a very large quantity of hydrogen and ammonia, leaving much valerianic acid in combination with the potash, and likewise leucine and tyrosine (Liebig).<sup>3</sup> When decomposed with chromic acid, or with sulphuric acid and binoxide of manganese, casein yields much more acetic acid, oil of bitter almonds, and benzoic acid, but much less valerianic acid and butyric acid than fibrin; in reference to the quantities of these products of decomposition it most nearly resembles albumen, although it yields a larger amount of acetic acid (Guckelberger).<sup>4</sup>

Simon has directed attention to certain differences presented by casein from *women's milk*, cows' milk, and the milk of the bitch. Casein from women's milk is white or yellowish, friable, becomes moist on exposure to the air, is insoluble in alcohol, but dissolves in water, forming a turbid, frothy fluid, from which it is completely thrown down by tannic acid, acetate of lead, and corrosive sublimate, and imperfectly precipitated by acetic acid and alum. Casein from *cows' milk* is not so freely soluble in water, and, when dry, is tough and horny; while that from the *milk of the bitch* is not tough and horny, and is difficult of solution in water. Dumas has, however, ascertained that the composition of these three kinds of casein is perfectly identical. There is much here that requires explanation. Simon's observations are certainly correct; and can not only confirm his statements from my own experience, but also those of Elsässer, according to which the cheesy coagulum of women's milk is always loose and jelly-like in its texture, while that of cows' milk is very firm and clotty. These differences may, however, be found to depend on many external relations, on the admixture of various substances, &c. Thus, for instance, I believe that the jelly-like coagula of women's milk are more dependent on the alkaline state of the fluid than on any peculiarity in the casein; at all events, I have found that women's milk, when acid, yields a much thicker coagulum than when alkaline, and cows' milk, when alkaline, a much looser coagulum than when acid;—facts of the highest interest and value in relation to dietetics.

*Composition.*—Casein, like albumen, has very often been analyzed, but all these analyses have led to no perfectly certain empirical formula, and far less to a rational one. We give as examples, analyses by

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 55, S. 78-95, and Bd. 58, S. 264-273.

<sup>2</sup> Handwörterb. der Chemie v. Liebig, Wöhler u. Pogg. Bd. 3, S. 220.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 57, S. 127-129.

<sup>4</sup> Ibid. Bd. 64, S. 89-100.

	Mulder. <sup>1</sup>	Schärer. <sup>2</sup>	and Dumas. <sup>3</sup>
Carbon, . . . . .	53.83 . . . . .	54.665 . . . . .	53.7
Hydrogen, . . . . .	7.15 . . . . .	7.465 . . . . .	7.2
Nitrogen, . . . . .	15.65 . . . . .	15.724 . . . . .	16.6
Oxygen, } . . . . .	28.37 . . . . .	22.146 . . . . .	22.5
Sulphur, }			
	<hr/> 100.00	<hr/> 100.000	<hr/> 100.0

According to more recent investigations purified casein contains 0.85% of sulphur.

In casein, precipitated by acetic acid, and washed with alcohol and ether, Rüling<sup>4</sup> found 1.015% of sulphur; but in casein which had been precipitated by acetic acid, dissolved in carbonate of soda, and again precipitated by the acid, the quantity was only 0.850%; Walther<sup>5</sup> found 0.933%, and Verdeil<sup>6</sup> 0.842% of sulphur in casein, which had been treated with hydrochloric acid and carbonate of soda.

According to Mulder, casein is nothing more than his hypothetical protein combined with sulphamide. No formula for casein can, however, be established till the question is definitively settled whether it be a simple or a compound body.

Casein that has not been treated with acids contains about 6% of phosphate of lime; more, consequently, than is contained in any of the protein-compounds we have hitherto considered.

*Preparation.*—We obtain soluble casein by evaporating skimmed milk, extracting the residue with ether, and dissolving it in water; we then throw down the casein from the aqueous solution by the addition of alcohol, with which we must also carefully wash the precipitate.

Berzelius precipitates the casein from skimmed milk by sulphuric acid, rinses the white coagulum with water, and decomposes the sulphate of casein with carbonate of lime, or (which is better) with carbonate of lead; the casein which is dissolved in water always contains a little lead, which, however, may be removed from the solution by sulphuretted hydrogen.

Simon removed the fat, by means of alcohol and ether, from casein precipitated by sulphuric acid, before decomposing it with carbonate of lime.

Mulder prepared casein for elementary analysis by precipitating it from skimmed milk, by warming it with acetic acid, washing and thoroughly rinsing the precipitate with water, separating the fat by boiling alcohol, and finally, by drying at 130°.

According to Rochleder's<sup>7</sup> method skimmed milk is coagulated with dilute sulphuric acid (acetic acid or hydrochloric acid may however be used in its place); the precipitate is then duly pressed and again dissolved in a dilute solution of carbonate of soda; this solution is allowed to stand for some time in a shallow vessel, when there gradually forms on its surface a layer of fatty matter, which we must remove as completely as possible with a spoon, or else we must decant the subjacent fluid with a syphon. The fluid is now again precipitated with an acid,

<sup>1</sup> *Bullet. de Néerl.* 1839, p. 10.

<sup>2</sup> *Compt. rend.* T. 21, p. 715.

<sup>3</sup> *Ibid.* Bd. 37, S. 316.

<sup>4</sup> *Ibid.* Bd. 46, S. 253-256.

<sup>5</sup> *Ann. d. Ch. u. Pharm.* Bd. 40, S. 40.

<sup>6</sup> *Ann. d. Ch. u. Pharm.* Bd. 38, S. 300.

<sup>7</sup> *Ibid.* Bd. 38, S. 319.

and the previous steps are repeated. After the casein has been thrice dissolved in carbonate of soda, and the fat as often skimmed off, the last trace of fatty matter may then be easily removed by alcohol and ether, which otherwise is a very difficult task. Casein thus prepared may moreover be rendered entirely free from acid by repeated boiling in water; so that if, for instance, it has been precipitated with sulphuric acid, chloride of barium does not excite the slightest turbidity when added to its acid solution. Bopp<sup>1</sup> adopts a modification of Rochleder's method; he precipitates a solution of casein in carbonate of soda with hydrochloric acid, and repeatedly washes this precipitate with water containing 2% or 3% of hydrochloric acid; it is then mixed with pure water, in which it swells and gradually dissolves, especially if the temperature be raised to about 40°; the solution contains hydrochlorate of casein, from which the casein must be thrown down by careful neutralization with an alkali, and the precipitate then washed.

*Tests.*—It is now ascertained, that no reliance is to be placed on certain properties of casein which were formerly regarded as characteristic indications of its presence, and it is unfortunately the case that recent investigations have only shown us the fallacy of our former tests, without affording us better and more certain means of detecting it. There were three especial properties by which it was generally believed that casein might be recognized. In the first place the capability of an animal fluid to form a membrane on evaporation, was regarded as the most certain sign of the presence of casein; we have however already shown (p. 297) that both alkaline albuminates and acid solutions of albumen equally possess this property, and, indeed, that the fluid filtered from ordinary coagulated albumen always contains such an albuminate, and consequently has a tendency to form such a membrane; the tendency of an albuminous fluid to form a membrane on evaporation, is directly proportional to the amount of alkali or albuminate which it contains, and it is this circumstance that has led some very accurate observers to believe that they have found casein in the blood and in fluid exudations, where in reality not a trace of this substance occurs.

[Since the publication of this volume in German, two memoirs on the assumed discovery of casein in the blood have appeared, one by Guillot and Leblanc,<sup>2</sup> the other by Panum.<sup>3</sup>—G. E. D.]

This error would be further promoted by a second mode of testing for casein, namely, by its property of being precipitated by acetic acid; this was regarded as a means of distinguishing between casein and albumen; but if the slight turbidity which affects albuminous solutions (see p. 296), when they are neutralized or very much diluted with water, occasionally gave rise to a confusion between these substances, this must have occurred far more frequently when it was believed that the albumen had been removed by boiling from albuminous fluids; for there then remains, as we have already seen, a little coagulated albumen with soda or potash in solution; by the addition of acetic acid the albumen is precipitated from the solution in precisely the same manner as casein, which is not the case with the unboiled albuminate of potash. Every accurate

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 69, S. 16-37.

<sup>2</sup> Compt. rend. T. 31, p. 585.

<sup>3</sup> Arch. f. pathol. Anat. Bd. 3, S. 251.

experimenter must have thus been led (till these facts were ascertained) to believe that he had always found a little casein in the fluid filtered from coagulated albumen.

The third means of discovering casein is the only one now left us; and even this, by its incorrect application, has already given rise to false conclusions. We refer to the *coagulability of casein by rennet*—a test by which some have supposed that they have detected casein in the blood: but in order that the casein may be separated by this means, the rennet must be tolerably fresh, or at all events must not have become putrid, when it is placed in the fluid which is to be examined; the mixture should then digest, at a temperature of  $40^{\circ}$ , for a period not exceeding two hours; if no coagulum is then formed, we are not justified in assuming that casein exists in the fluid; for if we allow the rennet to remain for twenty-four hours or longer in the fluid at that temperature, putrefaction ensues, with the development of *vibriones*, and the fluid becomes turbid by the products of putrefaction, but not by coagulated casein. Blood in which, for instance, some chemists fancy that they have thus detected casein, putrefies, on the addition of rennet, after a considerable time, but I have never succeeded in obtaining from it a true coagulum of casein.

*Sulphate of magnesia* and *chloride of calcium* have been recently recommended as very good tests for the presence of casein; the casein separating on *boiling* in combination with magnesia or lime; but unfortunately albuminate of soda (which, as we know, does not coagulate on boiling) possesses this property in common with casein.

At an earlier period of organic chemistry, many other reactions by which casein was characterized used to be described, as, for instance, sulphurous acid, its difficult solubility in acetic acid, &c.; but all these means yield no definite result. Moreover, during the last few years, much attention has been devoted to the behavior of casein and of the protein-compounds generally with tests of the most varied kind; but however deserving of notice such endeavors may be, they have not produced any great results, nor indeed could they be expected to do so, for independently of the fact that an endeavor to discover any decisive reactions is mere groping in the dark, when the investigation is not guided by one uniform leading idea, the results of these experiments so frequently vary in their individual character that it is often impossible to bring them into harmony. Any one who has occupied himself with such investigations, and observed the action of acids, bases, metallic salts, &c., under various relations, on the albuminous substances, can confirm the statement that one and the same substance, under apparently similar relations, yields the greatest diversity of reactions, sometimes presenting a similarity to one and sometimes to another protein-compound. The various relations which modify these reactions, and of whose nature we are still ignorant, render experiments perfectly useless, unless these circumstances be taken into account. In general we may suspect the modifying influence, but in special cases we are often quite in the dark. A very simple example will illustrate our meaning. Casein is sometimes very readily soluble in acetic acid, at other times it is rather difficult of solution, while again there are other occasions in which it is almost insoluble in

that fluid; we can only conjecture that the state of cohesion, the earthy matters contained in it, &c., give rise to this difference; but in individual cases it is often impossible to say which of these two conditions, or whether any other, is influencing the result of the special observation.

I may in this place give another example of the difference produced by inexplicable circumstances on reactions: on one occasion a turbid acid solution of casein becomes perfectly clear on the application of heat, on another the casein is entirely separated on heating; and thus acetic acid not unfrequently produces only a slight precipitation in the milk of cows and other animals, a true coagulum only separating on boiling.

In order to determine with any certainty whether casein exists in an albuminous fluid, we should conduct our experiment in the following manner. The fluid must be boiled for some time, a little hydrochlorate of ammonia having been first added, to effect the separation of the albuminate of soda; we must then filter it, and ascertain whether sulphate of magnesia or chloride of calcium yields a precipitate without the aid of heat; if such a precipitate be formed, we remove it by filtration, before boiling the fluid, in order to search for casein. If a precipitate be formed on boiling the fluid thus prepared, the presence of casein must in this case be shown by rennet.

Acetic acid was formerly almost the only reagent employed in the *quantitative determination* of casein; but this acid by no means effects a thorough precipitation of the casein, and when added in excess it often dissolves a very considerable portion;—an observation which formerly led Schübler to the belief that the milk contained a peculiar substance, to which he gave a special name, *zieger*.<sup>1</sup> The best method of analyzing milk which has yet been proposed is, unquestionably that of Haidlen.<sup>2</sup> On stirring milk with about one-fifth of its weight of finely pulverized gypsum, and heating it to 100°, a perfect coagulation ensues, and we obtain on evaporation a brittle, easily pulverizable residue, from which ether and alcohol easily remove the fat milk-sugar, and most of the salts. The residue is then not pure casein, but the quantity of that ingredient in a state of purity may be easily calculated by determining the quantity of fat, sugar, and salts contained in the milk.

### *Physiological Relations.*

*Occurrence.*—Casein occurs, as is well known, in the milk of all the mammalia.

Clemm<sup>3</sup> found 3·37%, and Fr. Simon,<sup>4</sup> on an average, 3·5% of casein in *women's milk*; the latter found 4% in the colostrum, but only 2·15% in the milk six days after delivery. In women's milk of good quality Haidlen<sup>5</sup> found 3·1%, but in milk of an inferior character only 2·7%.

In *cows' milk* Boussingault<sup>6</sup> found the casein to range from 3% to 3·4%. Playfair determined the average at 4·16%, Poggiale<sup>7</sup> at 3·8%, and Simon at 7%.

In the *milk of bitches* Simon found 14·6% of casein, Dumas<sup>8</sup> from

<sup>1</sup> [Zieger is, literally, a sort of whey.—G. E. D.]

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 45, S. 273 ff. <sup>3</sup> Inquis. chem. etc. Götting. 1845.

<sup>4</sup> Frauenmilch. Berl. 1838.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 45, S. 273 ff.

<sup>6</sup> Ann. de Chim. et de Phys. 3 Sér. T. 8, p. 94.

<sup>7</sup> Compt. rend. T. 18, pp. 503–507.

<sup>8</sup> Ibid. Bd. 21, S. 708–717.

9.73% to 13.6%, and Bensch<sup>1</sup> from 8.34% to 10.24% (including the insoluble salts). In asses' milk Peligot<sup>2</sup> found 1.95%, and Stiptr. Luiscius and Bondt<sup>3</sup> 2.3%; the latter found 16.2% in mares' milk; in goats' milk, Payen found 4.52%, Stiptr. Luiscius and Bondt 9.12%, and Clemm 6.03%; Schlossberger<sup>4</sup> found 9.66% in the milk of a he-goat, and Stiptr. Luiscius and Bondt 15.3% in ewes' milk.

According to Dumas and Bensch the milk contains more casein during an *animal* than during a strictly *vegetable* diet.

The nitrogenous substance to which we apply the name of casein, occurs in the milk, for the most part, in a state of solution, but a not inconsiderable portion forms the free investing membrane or wall of the milk-globules. The microscope alone affords us no information regarding the structure of this membrane; hence we do not attach much faith to the assertions of Raspail and Donne,<sup>5</sup> who were the first to assume the existence of such a membrane: Simon<sup>6</sup> believed that he had detected fragments of these membranes in milk which had been evaporated and treated with ether; Henle<sup>7</sup> was the first to demonstrate its existence; on examining under the microscope the gradual action of acetic acid on the milk-globules, he noticed a decided distortion of this membrane. The best proof of the existence of an investing membrane is, however, afforded by an experiment instituted by E. Mitscherlich: on shaking perfectly *fresh* milk with ether, it is scarcely at all changed, the ether merely taking up a little fat; now, if the milk were a simple emulsion, it would yield all its fat to the ether, and would be converted into a transparent, tolerably clear fluid; as this is not the case, the separate fat-vesicles must be surrounded by an insoluble substance; if now we add a substance capable of dissolving these membranes, ether when shaken with milk will act on it precisely as on an emulsion, that is to say, it will take up the fatty matter; and indeed this is the case if a little caustic or carbonated alkali be added to the milk before it is shaken with ether. Mitscherlich, by this beautiful experiment has removed all doubt regarding the existence of such a membrane. I have, however, observed the following facts: on placing under the microscope milk shaken with ether but to which no potash has been added, the surface of the milk-globules appears of diminished transparency, opaque, and fissured; in short, the wall presents the appearance of being coagulated. In place of potash I have used phosphate of soda and sulphate of soda; milk, treated with the former, yielded almost all its fat to ether, but did not become so clear as when treated with potash; under the microscope the aqueous fluid exhibited only a few fat-globules, which were *no longer round* but corrugated, of a caudate form, &c. Sulphate of soda has the property of causing the capsules of the milk-globules to burst, after which the fat can be extracted from the milk by ether; the watery fluid, however, remains very turbid, but no longer exhibits under the microscope either milk-globules, or shreds of destroyed capsules, but only

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 221-227.

<sup>2</sup> Ann. de Chim. et de Phys. T. 62, p. 432.

<sup>3</sup> Mémoires de la Soc. de Méd. de Paris, 1787, p. 525.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 51, S. 431.

<sup>5</sup> Cours de Microscopie, p. 356.

<sup>6</sup> Medic. Chem. Bd. 2, S. 75, or English Translation, vol. 2, p. 48.

<sup>7</sup> Fror. Notiz. 1839, Nr. 223, and Allgemeine Anatomie, S. 942.

extremely minute, scarcely isolable, molecular granules, which are unquestionably the fragments of the destroyed capsules, and do not consist of finely comminuted fat; for, on the addition of a little potash, they not only do not disappear under the microscope, but the fluid which had previously retained its milky color becomes perfectly clear and limpid. Hence we perceive that our ordinary casein not only contains the protein-compound dissolved in the milk, but likewise another, which forms the capsule of the milk-corpuscles, so that we thus also have a microscopico-mechanical proof of the composite nature of ordinary casein.

From a comparatively early epoch in animal chemistry attempts have been made to recognize casein in the blood; but none of them were distinctly successful. Recently, however, very careful investigations have been made by several different persons, as for instance, by Guillot and Leblanc,<sup>1</sup> Panum,<sup>2</sup> and Moleschott,<sup>3</sup> which demonstrate the existence of a substance in the serum which appears to be different from the ordinary albumen, and which they hold to be identical with casein. Whether this substance is to be regarded as perfectly identical with ordinary albumen (as Scherer and I hold), the difference in its properties depending only on certain admixtures or incidental relations, is a point that possibly may never be decided; this much, however, is certain, that although the presence of casein in the blood is *a priori* in the highest degree probable (in consequence of its occurrence in other fluids), yet the identity of this constituent of the blood with the casein of the milk is by no means definitely established. Such questions as these can, however, never be thoroughly decided until we are better acquainted generally with the chemical constitution of the protein-bodies.

Guillot and Leblanc have obtained their casein by the addition of a few drops of acetic acid to blood-serum after the removal of its albumen by heat; and they maintain that they found in the precipitate all the characters of casein; they do not, however, state what these properties are. Anything like a doubt as to whether the substance precipitated by acetic acid was casein or albumen, or some other special substance, seems never to have occurred to these investigators.

The quantity of this substance precipitable by acetic acid was, according to their observations, different in different animals, and varies with the sex, food, bodily conditions, &c. It was especially abundant in the blood shortly before delivery, and during the process of lactation, the actual maximum occurring soon after delivery. In many pathological conditions this substance entirely disappeared from the blood.

The substance precipitable by acetic acid occurs, according to Stas,<sup>4</sup> in very large quantity in the serum from the blood of the umbilical cord and the placenta.

Panum considers that the precipitate mentioned in p. 296, which is obtained by the dilution of the blood, especially after the addition of a little acetic acid, and which Scherer regards as albumen poor in salts and free from an alkali, is casein; and he terms it *serum-casein*. On drying, this substance first becomes transparent and viscid, then glistening, hard, and tough, assuming, as Panum strongly urges, a beautiful

<sup>1</sup> Compt. rend. T. 31, p. 585.

<sup>2</sup> Arch. f. pathol. Anat. Bd. 8, S. 251-272.

<sup>3</sup> Arch. f. physiol. Heilk. Bd. 11, S. 105-111.

<sup>4</sup> Compt. rend. T. 31, p. 680.



green color. Scherer,<sup>1</sup> under whose direction Panum conducted his experiments, correctly remarks, that the differences between this substance and albumen depend more on the nature of the fluids in which they occur, on the weakened action of the salts, the great quantity of the water, and the extremely minute disintegration of the separated matter, than on an essential difference in the nature of this substance as compared with ordinary albumen, and that casein is precipitated from concentrated, as well as from dilute solutions, while this substance is only precipitated from very dilute solutions by acetic acid. Panum remarks as characteristic of this substance, that it is precipitated from its solutions by carbonic acid: this observation is quite correct, but it stands in direct opposition to the view, that this substance is identical with casein; for as far as my experience goes, the casein of milk is not precipitated by carbonic acid, although the globulin of the crystalline lens is almost entirely thrown down from its watery solution by carbonic acid. Moreover, this substance, which may also be recognized in small quantity in the white of egg, presents a much closer resemblance to globulin than to the ordinary casein of milk. Panum has also found more of this substance in the serum of woman's than in that of man's blood (0.3%); and it was especially abundant in the serum of women shortly after delivery (from 0.99 to 1.25%).

Although Panum's experiments were very carefully made, and have led to the discovery of many new facts, yet the far less numerous experiments of Moleschott, who treated the serum, after the removal of the albumen by salts and coagulation, with sulphate of magnesia and heat, seem to afford far stronger evidence in favor of the existence of casein in the blood. I will here repeat, that neither Scherer nor I have ever ventured to deny, that in all probability casein exists in the blood; but until its presence in that fluid is actually proved, we cannot recognize its existence there. The discussion on this point is, however, little more than a war of words, for how can we strictly identify a substance with casein when we do not know what casein actually is, or rather believe that it is a mixture of two or more substances?

M. S. Schultze<sup>2</sup> has found a matter coagulable in the cold by acetic acid in the interstitial juice of the middle coat of the arteries, and Moleschott<sup>3</sup> in that of the connective tissue, and of the ligamentum nuchæ; and I have found the same substance in all contractile tissues, which contain contractile fibre-cells (smooth muscular fibres).

Stas<sup>4</sup> found a similar substance in the fluid of the allantois.

It was formerly supposed that casein existed in other animal fluids and solid parts, and indeed it was regarded as a normal constituent of the blood. In our consideration of the means by which casein may be recognized with certainty, we have, however, shown that no reliance can be placed on statements of this nature. Hence we can attach no weight to the assertions that casein occurs in the urine or in effusions within the peritoneum, the pleura, or the arachnoid, and the cases where, in consequence of metastasis of the milk, casein actually occurs in the urine or

<sup>1</sup> Jahresber. d. ges. Med. 1851, S. 75.    <sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 71, S. 217.

<sup>3</sup> Physiol. d. Stoffwechsels. Erlangen, 1851, S. 366.

<sup>4</sup> Compt. rend. T. 31, p. 630.

black. On being heated, however, they become yellow and dissolve with tolerable readiness into a yellow fluid. The aqueous solution of the crystals yields a light-brownish flocculent precipitate, even when very much diluted.

*Hydrochloric* and *sulphuric acids* do not give rise to any precipitates from the watery solution of the tetrahedric crystals, although they precipitate the solution of the prismatic crystals: this difference depends, however, solely upon the different concentration of the solutions; for if the solution of the prismatic crystals be diluted, as for instance, by the addition of four times its volume of water, no precipitate will be formed either with hydrochloric or sulphuric acid; but if, on the other hand, from four to six times the volume of concentrated hydrochloric acid, or an equal volume of English sulphuric acid, be added to a solution of the tetrahedric crystals, this substance will likewise be precipitated.

The crystallizable substance is easily soluble in *acetic acid*, which simply changes the color of the red watery solution into a brownish-yellow. If we *neutralize* with ammonia the fluid which has been acidified with acetic acid, pale-brownish flakes are separated. Like other protein-bodies, the crystalline substance may be precipitated from the acid solution by yellow as well as by red prussiate of potash. It has also the further property in common with them, of being precipitated by neutral alkaline salts from the acetic-acid solution, or by acetic acid from the solution which has been treated with such salts. This precipitate which is thus obtained, is soluble in water, and exhibits very different properties from the original crystalline substance, a point to which we shall revert at a future page.

The crystals are insoluble in a concentrated *solution of potash*; they are, however, very readily dissolved by a dilute solution of potash as well as by *caustic ammonia*, when they exhibit a brownish-yellow color; this substance is precipitated from the alkaline solution by acetic acid in the form of light-brownish flakes, and this is the case even when the fluid exhibits only a faint alkaline reaction.

*Chlorine gas* decolorizes the solutions almost instantaneously, and precipitates white flakes.

An aqueous solution of *iodine* merely changes the red color of the fluid into a brownish-yellow.

The salts of the *alkalies* and the *alkaline earths* do not give rise to any precipitates.

*Nitrate of silver*, *bichloride of mercury*, *perchloride of iron*, *protochloride of tin*, and neutral and basic *acetate of lead*, do not yield the slightest reaction, and it is only when ammonia is added to the fluid, which has been treated with salts of lead, that a very voluminous and grumous precipitate is formed.

*Nitrate of protoxide of mercury* and *bichromate of potash* give rise to very considerable dirty white precipitates. Millon's test-fluid yields the reaction peculiar to all the protein-bodies.

*Sulphate of copper* leaves the fluid at first perfectly unchanged, but when it has stood for some time, it deposits an abundant pale-greenish precipitate.

A solution of pure crystals becomes gradually *decomposed* on exposure

to the air, although less rapidly than solutions which are mixed with other organic constituents of the blood. The crystals appear also to undergo a change when *dried in vacuo*, at all events their solution no longer presents the same bright red color. The crystals begin to decompose at a temperature of  $160^{\circ}$  or  $170^{\circ}$ ; at a higher temperature they swell considerably, and develope vapors which smell like burnt horn, and become strongly phosphorescent on being kindled: the substance is moreover readily consumed, leaving merely a small quantity of ash.

Alcohol renders the crystals insoluble in water, but it does not materially affect their shape—a remark which applies most forcibly to the tetrahedric form; the only change which they undergo being that their surfaces no longer appear perfectly plane; they remain nearly the same when heated to  $100^{\circ}$ . The coagulated crystals observed by Reichert<sup>1</sup> in the uterus of a pregnant rabbit were no doubt similar in character to these, for it is only the tetrahedra which, when treated with alcohol, exhibit all the remarkable properties which Reichert noticed in the crystals on which he made his observations. Thus, for instance, they swell in dilute acetic acid, so that their diameters are increased three or fourfold; but they recover their former volume when washed; or when the acid is neutralized. They must, therefore, be secondary crystals, formed from the coagulation of the originally soluble crystallized substance.

*Composition.*—The discovery of a crystallizable protein-substance appeared at once to afford a new means for obtaining more secure points of support for the establishment of its true constitution; but hitherto the elementary analyses of this substance have not furnished the desired information,—on the one hand, because the results obtained were too nearly identical with those yielded by the other protein-bodies, and on the other hand, because no guarantee of the perfect purity of the substance could be obtained. We must defer to a future page the consideration of the reasons which lead us to reject the validity of the results of former elementary analyses, and we will here only observe, that the membranes of the colored blood-corpuscles and the colored blood-cells penetrate through all filters and follow the blood-crystals, so that only a tolerably pure, and not an absolutely pure crystalline substance, can be obtained. In the mean while, we may at least hope to obtain a somewhat more definite insight into the constitution of this substance through its products of decomposition than we can possibly hope to attain in the case of the other protein-bodies. We have already mentioned that the different forms of the crystals of certain kinds of blood clearly show that the substances we have here to consider are homologous bodies, whose comparative analyses promise to afford at least some information regarding the constitution of these mysterious substances.

I have hitherto only analyzed this substance from the blood of guinea-pigs, and hence I cannot venture to found any conclusion on such analyses; both tetrahedric crystals and the prismatic (those of the dog) are very poor in ash-constituents: I found that both kinds contained about 1% of mineral substances, the principal part of which consisted of oxide of iron, which frequently amounted to 72% of the ash; about 21%

<sup>1</sup> Müller's Arch. 1849.

of the ash was phosphoric acid, while there was, moreover, a little lime and potash. This substance contained much less sulphur than is found in any other protein-substance.

As these crystals are always colored, the question here suggests itself, whether a special pigment (whose product of metamorphosis might be the well-known hæmatin, see p. 267) is here merely added to the true crystalline substance, and either crystallizes with this substance as an isomorphous body, or only colors it in the same manner as uric-acid crystals are commonly colored by the coloring matter of the urine, or whether we are here considering only a single ferruginous, crystallizable substance, of which hæmatin constitutes one of the separated products. I have not yet been able decisively to determine this question, but several facts seem to me to afford the greater amount of probability to the latter of these views.

*Products of its metamorphosis.*—These substances have not yet been analyzed with any satisfactory amount of exactness; we will therefore simply observe, that this protein-substance, precisely in the same manner as albumen, after being treated with acetic acid and alkaline salts, yields a substance which is altogether analogous with Panum's *acid albumen*. The aqueous solution of this substance does not exhibit the slightest turbidity on boiling, but when a larger or smaller quantity of an alkaline salt is added to it, a precipitate will be formed at a lower or higher temperature, precisely the same as in the case of acid albumen. An excess of salt precipitates this substance, even at an ordinary temperature; hence we may obtain it entirely free from acid, after repeated solution in water and precipitation by salts. When the solution containing an acid is neutralized by potash or ammonia, a considerable deposit is formed, which dissolves in ammonia, but is precipitated from it at a gentle heat. Nitric and sulphuric acids throw down copious precipitates from the aqueous solution, but hydrochloric acid does not produce such an effect. Ferrocyanide of potassium occasions a considerable deposit without any special addition of acid. Sulphate of magnesia, alum, sulphate of copper, chloride of iron, protochloride of tin, and neutral acetate of lead, do not produce any precipitates even by boiling, but precipitates are thrown down by basic acetate of lead, nitrate of silver, bichloride of mercury, and nitrate of protoxide of mercury.

I am still engaged in the analysis of this substance, as well as in the investigation of other products of decomposition of the crystalline substance.

*Preparation.*—The crystals of the blood, which may certainly have been seen by many earlier investigators, but which were first observed by O. Funke,<sup>1</sup> were prepared exclusively for microscopical examination by him and by F. Kundt,<sup>2</sup> to whom we owe the discovery of the tetrahedric and the hexagonal blood-crystals; the method they employed was, to cover a minute drop of blood with a glass slide, and after a small quantity of water, alcohol, or ether, had been poured upon it, the whole was exposed to gradual evaporation. I have now succeeded,<sup>3</sup> by different

<sup>1</sup> Dissert. inaug. Lips. 1851; and Zeitschr. f. rat. Med. N. F. Bd. 1, S. 148–192, Bd. 2, S. 199–244, u. 288–292.

<sup>2</sup> Zeitschr. f. rat. Med. N. F. Bd. 2, S. 271–287.

<sup>3</sup> Ber. d. k. sächs. Ges. d. Wiss. 1852, S. 23–26, u. 78–84.

methods, in exhibiting these crystals on a large scale and with tolerable quickness, and in all these modes of preparation light and atmospheric influences constitute the most essential conditions towards the rapid formation of these crystals. The method of preparation frequently requires to be very considerably modified in different kinds of blood. Funke has shown, in his careful experiments on the mode of formation of these crystals under a glass slide, that it is essentially necessary that the blood-cells should first burst before crystallization can begin, and the only available means are water, alcohol, and ether, as has been shown by Funke and Kunde. The evaporation which occurs after the formation of the crystals under a glass slide, is by no means so important as it would appear, since, for instance, the blood (of guinea-pigs) may be diluted with twice its volume of water, and yet the crystals may be perfectly separated in the course of three-quarters of an hour after the employment of a proper method of treatment; in other soluble crystals, as, for instance, in those of the dog, it is necessary to facilitate their separation by the addition of an adequate amount of alcohol.

*Tests.*—Although this substance differs so essentially from all other protein-bodies by its capacity for crystallization, its indifferent behavior towards moderately diluted hydrochloric and sulphuric acids, towards nitrate of silver, neutral and basic acetate of lead, bichloride of mercury, &c., it is extremely difficult and sometimes even impossible to recognize it, when it is present only in small quantities, or when it is mixed with many other protein-substances. Since other protein-bodies, or their immediate products of metamorphosis share at least in some of the properties which appertain to it, its presence in a mixture of protein-substances, could not be regarded as thoroughly proved, until its crystals had been obtained. But is it not probable that all the protein-bodies, or a substance separated from mineral matters and common to all of them, may crystallize? But even when crystals have actually been obtained from an albuminous fluid, it requires a very careful investigation to prove their identity with the crystalline substance of the blood.

#### *Physiological Relations.*

We have already remarked in the preceding pages, that the crystallizable substance of the blood is limited to the colored blood-corpuscles, as Funke has especially shown to be the case. It would appear, however, from experiments made on the subject, that it occurs in all red-blooded animals, although it may present the various modifications which have already been noticed; it is also more readily obtained from certain kinds of blood than from others.

We must yet enter somewhat more circumstantially into the mode of preparation of the crystallizable matter, since this subject is one of importance, when considered in reference to many still doubtful points referring to the blood. The blood-crystals are formed from blood containing fibrin and serum, as well as from blood which has been deprived of its fibrin, and possibly also from cruor freed from serum. Under certain relations, they are formed so rapidly and in such great quantities, that they frequently appear where one would the least expect to meet with them. Thus, for instance, they occur in great abundance

whenever blood-clots (as, for instance, from men, cats, and dogs) which have only been roughly chopped, and which have been frequently, although imperfectly washed in water, are suffered to remain for some time exposed in a moist state to the air, either in ordinary light, or, what is better, in sunlight; when thus treated, the superficial parts of the pieces of fibrin are rapidly covered with entire crusts of the most beautiful and large crystals. I obtained the tetrahedric crystals, to which I have already referred, most rapidly, that is to say, in 35 minutes after the animal had been killed, from the blood of guinea-pigs; the defibrinated blood, after being diluted with water and treated in the manner described in the preceding page (an aqueous extract of the cruor may also be employed for this purpose), is exposed for 15 minutes to a stream of oxygen either in broad daylight or sunlight, and carbonic acid is then conducted through the lighter red fluid for five, or at most ten minutes; the carbonic acid gradually renders the fluid darker, but it soon becomes more and more turbid; in accordance with the degree of its turbidity, the fluid exhibits a more or less bright vermilion-red tint from the separated crystals, which, when the stream of carbonic acid is interrupted, gradually sink to the bottom, and form a considerable bright vermilion-colored sediment. Much the same method must be employed to obtain the prismatic crystals from human blood, or the blood of cats and dogs; but in this case it is necessary to have recourse to several other conditions, which will subsequently be noticed. These crystals may, indeed, be separated by rinsing from all the constituents of the serum, and from the greater part of the colorless blood-corpuscles, as well as from the cell-membranes of the colored corpuscles, but still, notwithstanding repeated rinsings, many of the latter frequently remain, in consequence of having served, to a certain extent, as points of deposit for the crystals which thus enclose them; and hence they are not adapted, when in this condition, for elementary analysis. They must, therefore, be dissolved in water, and carefully filtered, in order perfectly to free them from all morphological particles. The recrystallization, however, presents great difficulties. We will here merely observe, that we cannot employ a high degree of heat on account of the coagulability of the substance, or the air-pump on account of the amount of gas necessary for crystallization. We may, moreover, recognize that the solution before us is that of a pure crystalline substance, from the fact that it cannot be precipitated by bichloride of mercury, nitrate of silver, or basic acetate of lead. The coagulum, which is obtained by heat from the crystalline solution, is at all events so far unsuited to elementary analysis, that it does not represent the pure crystalline substance; for during coagulation, the crystalline substance loses not only carbonic acid and phosphates, but also about 1.2% of organic matter, which consists of a strongly reacting acid and of a nitrogenous body, bearing some remote resemblance to gluten.

The numerous and variously modified experiments which I have instituted on this subject, lead me to regard light merely as an auxiliary in the crystallization; for, although crystals are certainly also formed in the dark, or even in the night under otherwise similar conditions, they are only gradually deposited, and always in far smaller quantities; thus, for instance, I could never obtain more than 2% of crystals from the

blood of the guinea-pigs in the dark, whilst I was frequently able to procure more than 7% of dry crystalline substance during ordinary daylight, or in sunlight. That which has been already stated in reference to light, applies very nearly with equal correctness to the application of oxygen. We may not unfrequently succeed, even without the use of oxygen, and by the mere application of carbonic acid, in obtaining these crystals in sunlight; but then only in far smaller quantities than in those cases in which the blood had been previously impregnated with oxygen. I discovered, from a series of comprehensive quantitative determinations, the particulars of which I have elsewhere<sup>1</sup> given, that the crystals are formed with far the greatest rapidity when the oxygen is suffered to pass through the blood in a slow stream for about 15 minutes before the application of the carbonic acid; for if carbonic acid be first, and oxygen be subsequently passed through the blood, the latter appears to hinder the process of crystallization; but when the fluid is introduced into carbonic acid after it has been impregnated with oxygen, the crystallization begins almost instantaneously. This crystallizing process appears, moreover, to occur with a rapidity proportional to the length of time that the fluid has been in contact with the oxygen before the application of the carbonic acid.

Different microscopical observations have appeared to show that the presence of fibrin is inimical to the formation of crystals, and that serum is indispensable to their production, but, as we have already observed, the presence of fibrin exerts no action, either favorable or the reverse, on the crystallization. The serum is equally devoid of all influence on this process, for crystals, and some very pure ones, may even be obtained from the later rinsings of chopped blood-clots, after they have been stirred and washed three or four times with water, although they certainly cannot retain any great amount of serum. No crystals, bearing even a remote affinity to the above-described blood-crystals, can be obtained from the serum either by these means, or by microscopical treatment under glass plates; hence we are scarcely going too far when we assert that observers who, like Robin, assert that they have procured the true blood-crystals from the serum, are entirely mistaken, and that they would be perfectly correct in regarding such crystals, which were noticed by every careful observer long before the discovery of the true blood-crystals, as mineral salts.

Although I very reluctantly enter upon the discussion of a subject which is still being made the object of inquiry, and cannot therefore be determined pending such an examination, I have thought that I could scarcely any longer avoid giving some notice of it. The observations to which I have already referred, together with others, incline me to believe that this crystalline substance is not a mixture of a pigment and a protein-body, but a pure chemical compound; the difference in the form of the crystals of different kinds of blood seems to indicate with tolerable certainty that this compound must, however, be either a salt-like or a conjugated compound. All the analyses which I have hitherto made of the pure substance have failed, like all previous elementary analyses of the protein-bodies, in yielding any definite views as to the constitution of

<sup>1</sup> Ber. d. königl. sächs. Ges. d. Wiss. zu Leipz. 1853.

this substance, but it seems to me that its recognition is rendered very simple on the supposition of a conjugation; the principal object to be had in view is, therefore, to discover some agent which will dissolve this conjugated compound, and separate the substance into its adjuncts; in how far I have succeeded in this purpose, I am scarcely able to determine. If the somewhat irrelevant question were asked, whether the crystalline substance is contained as such in the blood-corpuscles, existing in it only in a dissolved form, I could not directly affirm that such is the case, for the influence of such forces as light and oxygen, which are necessary to the formation of crystals, is inconceivable without the co-operation of chemical action: hence we might be led to assume that an oxidation had previously taken place. As, however, crystals cannot be formed without the co-operation of carbonic acid, mere oxidation cannot constitute the sole form of metamorphosis of the substance, for carbonic acid must essentially contribute towards the production of the new substance, which is then first rendered crystallizable. It might naturally be supposed that the investigation of this subject would enable us to decide the much-disputed question of the interchange of gases in the circulating blood, but the decision of this point is by no means so easy as we might be disposed to assume; at all events, owing to the small quantity by weight which is taken up by this substance, I have hitherto been unable to obtain any reliable results from my own quantitative determinations; other essential obstacles, moreover, hinder the determination of the gas which is to be absorbed. It must, moreover, be borne in mind that this capacity of the crystalline substance to be changed by the action of oxygen and carbonic acid is not peculiar to this body alone, but pertains without exception to nearly all the protein-bodies, as indeed every careful observer must have noticed, and as I have myself observed in the case of albumen, casein, globulin, &c., when submitted to a similar treatment with oxygen and carbonic acid. All protein-bodies undergo essential alterations in the open air, as has been observed in numerous instances (we need here only refer to the experiments of Scherer and Panum); but all persons who are conversant with such investigations must be aware of the extreme difficulty of tracing these metamorphoses, owing to the high atomic weight of these bodies. In the meanwhile, I am disposed to regard this crystalline substance as a combination with carbonic acid; and this view seems to derive confirmation, not only from its formation in a current of carbonic acid, and its spontaneous production in diseased liver and from putrefaction, but also from the incapacity of the solution to recrystallize after the dried or dissolved crystals have been placed under the air-pump; and finally, from that decided development of carbonic acid which we perceive in the moist crystals in vacuo, and the obviously more abundant development of gas in vacuo when acetic acid has been previously added to the solution. The globulin of the crystalline lens behaves in precisely the same manner, excepting that it is not crystallizable, and does not require the previous application of oxygen for its separation by carbonic acid. When a stream of carbonic acid is passed through the solution of globulin, the latter is precipitated, but this precipitate, on being shaken in pure water and in the open air, again dissolves into a clear fluid, from which the globulin may be again precipitated by carbonic acid. The crystalline substance which has been



treated with salt and acetic acid (corresponding to Panum's acid albumen) appears simply to undergo a metameric metamorphosis: it does not separate into several different substances on being coagulated by boiling (as Panum maintained was the case with albumen in the formation of acid albumen), but is rendered far more susceptible towards atmospheric influences than the original crystalline substance.

We now proceed to notice the chemical relations of certain substances which, perhaps, strictly speaking, do not belong to animal chemistry, since they occur only in the vegetable world: but there are two reasons, a chemical and physiological reason, why they should be noticed in the present place. In a chemical point of view they deserve notice, because we thus become acquainted with new protein-compounds, very similar to those already described, but yet differing from them, and thus obtain a more perfect insight into the whole group of this class of bodies: and in a physiological point of view they are of at least equal importance, for it is from them that the animal protein-compounds, which we have already described, are formed in the organisms of herbivorous animals, and that the solid substrata of the body are deposited in the various tissues. The actual physiological importance of these substances will be noticed when we enter upon the subject of "Nutrition."

### GLUTEN.

*Properties.*—This substance, to which the name *phytocolla* has also been applied, is, when dried, transparent, very hard and difficult to pulverize; when moist it is adhesive, viscid, and elastic; it is insoluble in cold, and very slightly soluble in hot water; it dissolves readily in boiling alcohol, from which water again precipitates it; it is also precipitated from its alcoholic solution by corrosive sublimate and acetate of lead; it dissolves imperfectly in acetic acid, and hence does not seem to be a perfectly pure protein-compound. In other respects it has all the properties of the protein-compounds.

*Composition.*—Gluten from several sources has been submitted to analysis; but here, as in the case of all the protein-compounds, no satisfactory formula has been calculated.

The following are the results of some of the analyses of this body:

	*	Scherer. <sup>1</sup>	Jones. <sup>2</sup>	Heldt. <sup>3</sup>	Mulder. <sup>4</sup>
Carbon, . . . . .		54.6	55.22	56.26	54.84
Hydrogen, . . . . .		7.4	7.42	7.97	7.05
Nitrogen, . . . . .		15.8	15.98	15.83	15.71
Oxygen, } . . . . .		22.2	21.38	19.94	21.80
Sulphur, } . . . . .					0.60
		100.0	100.00	100.00	100.00

The sulphur in gluten has been accurately determined by Rüling<sup>5</sup> and Verdeil;<sup>6</sup> the former found 1.134% in wheat-gluten and the latter 0.985% in rye-gluten.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 7.

<sup>2</sup> Ibid. S. 65–70.

<sup>3</sup> Ibid. Bd. 45, S. 191.

<sup>4</sup> Versuch einer allg. phys. Ch. 1844. S. 308.

<sup>5</sup> Ann. d. Ch. u. Pharm. Bd. 58, S. 310.

<sup>6</sup> Ibid. S. 318.

It is obvious that the numbers yielded by the above analyses differ too widely to admit of our attempting to calculate a trustworthy formula.

*Preparation.*—As this body especially occurs in the seeds of the cereals, the best method of obtaining it is by sufficiently kneading their flour under water, boiling the residue with alcohol in order to effect a perfect removal of the starch, and filtering while hot; on cooling and evaporating the solution, it is precipitated in white flocculi.

### LEGUMIN.

*Properties.*—This body forms either a white, nacreous, iridescent precipitate, or else is thrown down in a flocculent form; when dry, it has a yellow, transparent appearance, and is brittle. It coagulates like albumen from its aqueous solution, but is precipitated from it by acetic and phosphoric acid like casein, from which, however, it differs, in the first place, in not dissolving in concentrated acetic acid, and, secondly, in the circumstance that when it is precipitated by an acid, the precipitate does not dissolve when digested with carbonate of lime or of baryta. It is coagulated by rennet. It dissolves readily in ammonia and other alkalis.

*Composition.*—No definite results have as yet been obtained from the analyses of legumin. The following numbers have been found by the chemists whose names are attached to each analysis:

	Dumas & Cahours. <sup>1</sup>	Jones. <sup>2</sup>	Rochleder. <sup>3</sup>	Rüling. <sup>4</sup>
Carbon, . . . . .	50.50	55.05	56.24	50.59
Hydrogen, . . . . .	6.78	7.59	7.97	6.83
Nitrogen, . . . . .	18.17	15.89	15.83	16.54
Oxygen, } . . . . .	24.55	21.47	19.96	{ 25.57
Sulphur, }				{ 0.47
	100.00	100.00	100.00	100.00

The differences presented by these analyses are so great that it is obvious that we have not yet succeeded in obtaining this substance in a state of purity, and fit for elementary analysis.

*Preparation.*—This body is chiefly found in peas and beans, and other leguminous seeds, from which it may be easily obtained; the watery extract of these seeds has an acid reaction, and on neutralization the legumin is precipitated; it is purified by solution in ammonia, from which it is again precipitated by an acid, and finally by extraction with alcohol and ether.

Besides these substances, there are in the vegetable kingdom, and especially in seeds, other substances, which approximate more or less closely to the protein-compounds of the animal kingdom. In the first place there is *vegetable albumen*, which Liebig calls *vegetable fibrin*; it is insoluble in water, and similar in its composition to coagulated animal albumen; it remains undissolved, when we have separated the starch

<sup>1</sup> Ann. de Chim. et de Phys. T. 6, p. 409.

<sup>2</sup> Ibid. Bd. 46, S. 155.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 67.

<sup>4</sup> Ibid. Bd. 58, S. 301-315.

from flour by washing, and the gluten by alcohol. Of the *diastase* or *mucin* which is formed during the germination of grain, and which is a product of the metamorphosis of the previous substances, we know even less, both in reference to its composition and its properties. It appears from the investigations of Ortlöff<sup>1</sup> and Buckland W. Bull<sup>2</sup> that the *emulsin* or *synaptase* obtained from almonds is not a protein-compound; indeed this is sufficiently obvious from the large quantity of oxygen (26.56%) which it contains.

There are several animal substances pertaining to the protein-compounds of which we have no more accurate knowledge than we have of the above-named vegetable substances; in this category we may place *keratin*, the substance deposited in horny tissue (which, according to Mulder, is the same oxide of protein as exists in fibrin, but combined with a far larger quantity of sulphamide), the substance termed *mucin*, peculiar to mucus, and the *pyin*, existing in pus and morbid tumors, of which full notice will be taken when we treat of the chemical theory of the tissues and juices. In the same manner we shall treat of *pepsin* and the *peptones* when we enter into the special consideration of the digestive process.

## TEROXIDE OF PROTEIN (PROTEINTRITOXID).

### *Chemical Relations.*

*Properties.*—When dried, this substance is brittle, and easily pulverizable, but when moist it is tough, viscid, capable of being drawn out in threads, and when warmed has an odor resembling that of gelatin; it is soluble in water, but insoluble in alcohol and ether, and in the fatty and volatile oils; it has no reaction on vegetable colors. It is precipitated from its solution by dilute mineral acids, chlorine water, tannic acid, corrosive sublimate, the salts of the oxides of lead, silver, zinc, and iron, but not by ferrocyanide of potassium, the alkaline salts, or chloride of barium. With alkalis it forms neutral compounds, from which it is also precipitated by metallic salts. When boiled with caustic alkalis it develops ammonia, and becomes converted into a substance, which, according to Mulder, is the true teroxide of his protein, in accordance with his latest formula,  $C_{36}H_{25}N_4O_{10} + 3O + 3HO$ .

*Composition.*—This body was discovered and analyzed by Mulder;<sup>3</sup> from the mean of five analyses it was found to contain:

Carbon, . . . . .	51.69
Hydrogen, . . . . .	6.64
Nitrogen, . . . . .	15.09
Oxygen, . . . . .	26.58
	<hr/>
	100.00

In his most recent memoir Mulder regards this substance as a combi-

<sup>1</sup> Arch. d. Pharm. Bd. 48, S. 12-27.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 69, S. 145-162.

<sup>3</sup> Journ. f. pr. Ch. Bd. 22, S. 340; Bull. de Néerlande, 1839, p. 404; Ann. d. Ch. u. Pharm. Bd. 47, S. 300-320.

nation of true teroxide of protein with ammonia, in accordance with the formula  $\text{H}_4\text{NO} + 2 (\text{C}_{36}\text{H}_{25}\text{N}_4\text{O}_{13}) + 3\text{HIO}$ .

*Preparation.*—Mulder originally obtained this substance by treating his albumen-protein with chlorine, whereby he obtained the body which he then termed chlorite of protein; this substance when decomposed with ammonia yielded the body in question.

He subsequently ascertained that he could obtain it by the prolonged boiling of fibrin or albumen in water, if freely exposed to the air; the solution which is thus obtained is filtered and evaporated, and the residue extracted with alcohol; the portion insoluble in alcohol is again dissolved in water and precipitated by basic acetate of lead; the precipitate after being thoroughly washed is then decomposed by sulphuretted hydrogen, the sulphide of lead removed by filtration, and the solution evaporated.

*Tests.*—This body has so few characteristic properties, that in the present state of our knowledge it is extremely difficult, if not impossible, to distinguish it with perfect certainty from those substances which frequently occur, although only in small quantities, which have been hitherto named extractive matters soluble in water.

The peptones, ptyalin, pyin, and other little-investigated animal matters are very similar to this substance, but differ from it in some of their characters, and hence must not be regarded as identical with it, although many of the differences may be dependent on the admixture of other matters with them. Hence organic analytical chemistry has here a great blank to fill up in order to elucidate the actual conditions under which this substance occurs. Unfortunately it cannot be obtained in a state of purity from the animal fluids, so that we cannot have recourse to an elementary analysis to confirm our diagnosis.

#### *Physiological Relations.*

According to Mulder this body exists in normal blood and in all fluid exudations, and hence also in pus; and its quantity is very considerably increased in the blood in inflammatory diseases. He regards the *pyin* discovered by Güterbock in pus as altogether identical with this substance; but if for the reasons we have already given in reference to testing for teroxide of protein, we cannot regard it as positively decided that this substance occurs in all these animal fluids, yet it is probable from the mode in which it is artificially prepared, that a substance which is formed from albumen or fibrin in warm water exposed to the air, also occurs in the blood where the above-named substances which yield it, are exposed to similar influences. If more accurate investigations confirm the existence of this teroxide of protein in the manner that Mulder supposes, we shall then acquire a knowledge of an important intermediate link in the metamorphoses of the animal tissues, and in particular we shall have considerably approximated to the yet unsolved problem of the conversion of albuminous bodies into bodies yielding gelatin, or of fibrin into tissue.

## DERIVATIVES OF THE PROTEIN-COMPOUNDS.

The bodies of this group present very great differences in their physical and chemical properties; except that they all contain nitrogen, and that they occur only in the animal body, where they form the chief groundwork of the tissues, there is scarcely a point of general resemblance between them; in their behavior towards acetic acid and ferrocyanide of potassium, and towards concentrated hydrochloric and nitric acids they exhibit none of the essential characters of the protein-compounds. Only four of these substances have as yet been accurately studied, although regarding even their intimate chemical constitution there is as much doubt as in the case of the protein-compounds.

## ANIMAL GELATIN.

Under the term gelatin we comprehend those animal substances which do not exist ready formed in that state in the animal organism, but are produced from certain animal parts by mere boiling with water, so that the still undescribed substance from which this body is so easily obtained, may be regarded as the organic substratum of most of the animal fluids. All these very similar bodies, to which we give the common name of *gelatin*, are especially distinguished by the following properties; they swell and become very translucent in cold water; they dissolve in hot water; on cooling they separate as translucent, lubricous masses, and are precipitated from the most dilute solutions by chlorine, tannic acid, and most of the salts of the earths and metals.

There are two principal varieties of gelatin to be considered, namely, *bone-gelatin*, *carpenters' glue*, or *glutin*, and *cartilage-gelatin* or *chondrin*, although here, as in the case of protein, there appear to be several modifications of each variety.

## GLUTIN.

*Chemical Relations.*

*Properties.*—In a state of purity, glutin appears in colorless, transparent pieces, which are hard, horny, brittle, heavier than water, devoid of taste and smell, and exhibit no reaction on vegetable colors; on trituration it does not adhere to the pestle like the protein-compounds.

Glutin immersed in cold *water*, becomes soft, swells, and loses its transparency; in *warm water* it dissolves, forming a colorless, viscid solution, from which, on cooling, it separates as a jelly; Bostock's experiments show that good hard glutin will separate in this manner when diluted with 100 times its bulk of water. After being repeatedly dissolved in hot water, it loses the property of gelatinizing. Gelatinized glutin gradually becomes acid on exposure to the air, and then loses its property of fixing and binding. It is perfectly insoluble in alcohol,

ether, fats, and volatile oils; on the addition of *alcohol* to its warm solution, it coagulates into a white, tenacious, almost fibrous mass, which, however, readily dissolves again when warmed in pure water.

*Acids* and *alkalies* throw down no precipitate from aqueous solutions of gelatin; the latter in a dilute state precipitates a little bone-earth. Of the organic acids, *tannic acid* is the only one which throws down a precipitate from a solution of glutin; the precipitate is white and cheesy, and is observable even if the glutin be dissolved in 5000 times its weight of water.

The only *earthy* and *metallic salts* which precipitate glutin are corrosive sublimate, bichloride of platinum, and sulphate of binoxide of platinum. *Ferrocyanide of potassium* does not affect either its neutral or its acid solution. *Chlorine*, *bromine*, and *iodine*, on the other hand, act very powerfully on a solution of glutin; chlorine causes the separation of a coagulum which is partially thready, and after prolonged action, compounds are formed of chlorous acid and undecomposed glutin. *Creosote* gives a milky appearance to the clear solution; the salts of alumina, suboxide of mercury, the oxides of silver, copper, and lead, and of protoxide and peroxide of iron, exhibit no reactions when added to a solution of glutin, or, at most, cause only a very slight turbidity; and the same is the case with basic acetate of lead. *Basic sulphate of binoxide of iron* when added to a solution of glutin, causes a bulky precipitate, which, when dried, is of a deep red color.

Moist glutin exposed to the air soon undergoes putrefaction; it first becomes sour, but afterwards develops a large quantity of ammonia; according to Gannal,<sup>1</sup> the gelatinous tissues are the first of the solid animal structures to become putrid.

Dry glutin when *heated* softens, swells up, evolves an odor of burned horn, does not easily catch fire, and after burning for a very short time, leaves a voluminous, blistered, glistening coal, which after perfect combustion, yields a somewhat varying amount of phosphate of lime. The products of its dry distillation are those of the animal tissues generally; it yields, however, a preponderating quantity of carbonate of ammonia.

When boiled with concentrated *nitric acid*, glutin becomes gradually converted into oxalic and saccharic acids, and into two substances resembling suet and tannic acid. It dissolves in concentrated *sulphuric acid*, forming a colorless fluid, which on boiling gradually yields leucine, glycine, and other substances. If however it be treated with *sulphuric acid and peroxide of manganese or bichromate of potash*, it yields, according to Schlieper<sup>2</sup> and Guckelberger,<sup>3</sup> most of the non-nitrogenous acids of the first group ( $C_nH_{n-1}O_3$ ), and not only these but valerionitrile, hydrocyanic acid, hydride of benzoyl, benzoic acid, and certain aldehydes, and consequently precisely the same products of decomposition as the protein-compounds; it is, however, distinguished from them in yielding even less acetic acid than fibrin, very little benzoic acid and hydride of benzoyl, but on the other hand more valerianic acid than any of the protein-compounds.

<sup>1</sup> Hist. de l'embaumement, etc. Paris, 1838.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 59, S. 1-32.

<sup>3</sup> Ibid. Bd. 64, S. 39-100.

When boiled or fused with *hydrated potash* gluten developes ammonia, and is for the most part decomposed into leucine and glycine.

*Composition.*—Glutin has been analyzed by Mulder,<sup>1</sup> Scherer,<sup>2</sup> and Goudoever.<sup>3</sup> They found it to contain :

	Mulder.	Scherer.	Goudoever.
Carbon, . . . . .	50.40	50.76	50.00
Hydrogen, . . . . .	6.64	7.15	6.72
Nitrogen, . . . . .	18.34	18.32	—
Oxygen, . . . . .	24.62	23.77	—
	<hr/> 100.00	<hr/> 100.00	

No chemical formula that can be depended upon, has been deduced from these analyses. Mulder originally calculated  $C_{13}H_{10}N_2O_5$ , and Liebig  $C_{52}H_{40}N_8O_{20}$ , as the most correct formula. The calculations were for the most part based on its combinations with chlorous acid.

Schlieper<sup>4</sup> has found 0.12 to 0.14% of sulphur in gluten obtained from bones and ivory.

*Preparation.*—In order to prepare gluten in the purest possible form from common glue (which is obtained by boiling skins, tendons, &c., and the swimming-bladder of certain kinds of fish), Berzelius used to soften it in water, to expose it repeatedly to strong pressure, and then to suspend it in a linen bag in cold water till everything soluble in that fluid was removed. The softened gluten contained in the bag is then heated to 50°, when it becomes perfectly fluid, and must be rapidly filtered. The albuminous and mucous portions remain on the filter, while the hot solution of gluten passes through, and very soon again gelatinizes.

In order to prepare gluten from bones, we must digest them for a considerable time in dilute hydrochloric acid, in order to extract the bone-earth, allow the remaining cartilage to lie for some time in pure water in order to remove any adhering hydrochloric acid, and finally boil it with water. Gluten obtained from bones, skins, and tendons, has always a slightly yellow color.

Pure, colorless gluten can only be obtained from cellular tissue, shavings of hartshorn, calves' feet, and the swimming-bladder of certain fishes, by boiling them till they are thoroughly dissolved, filtering them while hot, and removing from them all foreign substances by the method recommended by Berzelius, which has been already described.

*Combinations.*—On passing *chlorine gas* into an aqueous solution of gluten, each bubble of gas becomes enveloped in a glutinous capsule; the fluid itself becomes milky; white flakes are observed on its surface, and at the bottom of the vessel we observe a deposit of a semi-transparent jelly. The substance which separates at the surface has a frothy, snow-white appearance, is tough and elastic, has a decided odor of chlorous acid, and can be dried at a temperature below 40° without becoming colored; after it has been partially dried, it may be deprived of all its water at 100°, and then no longer evolves any odor of chlorous acid. In this state the body is white, easily pulverizable, and insoluble both in

<sup>1</sup> Bullet. de Néerlande. T. 1, p. 23; Ann. d. Ch. u. Pharm. Bd. 46, S. 205–207.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 46–49.

<sup>3</sup> Ibid. Bd. 45, S. C2–C7.

<sup>4</sup> Ibid. Bd. 58, S. 379–381.

water and in alcohol. When ammonia is poured over it, nitrogen is developed, and hydrochlorate of ammonia and unchanged gluten are left.

Mulder<sup>1</sup> found that the action of chlorine and water on the organic substance gives rise to the formation of hydrochloric and chlorous acids, the latter of which enters into combination with the unchanged gluten, the compound consisting of 1 equivalent of acid and 4 equivalents of gluten.

Assuming that the composition of this substance is represented by the formula  $C_{52}H_{40}N_8O_{20} + ClO_3$ , its atomic weight = 8544.26. Mulder has found two other combinations of gluten with chlorous acid in the above-mentioned gelatinous deposit of the solution of gluten; one consisting of 1 atom of gluten with 1 atom of chlorous acid =  $C_{13}H_{10}N_2O_5 + ClO_3$ , and the other of 3 atoms of gluten and 2 atoms of acid =  $C_{39}H_{30}N_6O_{15} + 2ClO_3$ .

The action of *acids* on gluten has on the whole been as yet little examined; with dilute mineral acids it appears to enter into combinations, which, however, on cooling, gelatinize in the same manner as pure gluten. Concentrated *acetic acid* dissolves gluten which has been softened in water, and deprives it of the property of gelatinizing on cooling.

The only compound which has been carefully studied is that which it forms with *tannic acid*. This has been done by Mulder, who finds that, when freshly precipitated, it is white and curdy, when dried it is hard, brittle, and pulverizable, and that it is insoluble in water and alcohol. If the gluten is precipitated with an excess of tannic acid, we obtain a combination of equal equivalents of gluten and tannic acid =  $C_{13}H_{10}N_2O_5 + C_{18}H_7O_{11}$ ; if, on the other hand, there be an excess of gluten, the precipitate consists of 3 equivalents of gluten and 2 equivalents of tannic acid =  $C_{39}H_{30}N_6O_{15} + C_{36}H_{14}O_{22}$ .

No combinations of gluten with *alkalies*, *earths*, and pure *metallic oxides* are as yet known. Caustic lime dissolves in a solution of gluten. Gluten can, however, combine with several *basic salts*; a very considerable quantity of freshly precipitated bone-earth dissolves in a solution of gluten. Solutions of gluten, when treated with alum and with sulphate of peroxide of iron, do not yield a precipitate, except on the addition of an alkali; the precipitate in this case consists of gluten and a basic salt =  $Al_2O_3 \cdot SO_3$  or  $Fe_2O_3 \cdot 2SO_3$ . The precipitate obtained with sulphate of the binoxide of platinum appears to contain basic sulphate of binoxide of platinum =  $PtO_2 \cdot SO_3$ .

### *Physiological Relations.*

*Occurrence.*—Haller's remark: *Dimidium corporis humani gluten est*, now requires to be modified to the assertion that *half of the solid parts of the animal body are convertible, by boiling with water, into gelatin*; for actual gelatin is not contained in the animal organism. It has been for a long time maintained that gelatin is an actual constituent of the swimming-bladder of certain fishes; but even this is by no means probable. Scherer<sup>2</sup> has, however, recently found a substance in leucæ-

<sup>1</sup> Bull. de. Néerl. T. 2, p. 152.

<sup>2</sup> Verhandl. d. phys.-med. Ges. zu Würzburg. Bd. 2, S. 321-325.



mic blood which appears, from all its reactions, to be nothing else than glutin, and which consequently stands, in a chemical point of view, between the protein-bodies and gelatigenous matters.

It is, moreover, worthy of notice, that the embryo, up to the final period of its leaving the egg, contains no gelatigenous tissue (Hoppe).<sup>1</sup> Animal cell-walls and nuclei appear never to consist of gelatigenous tissue (Hoppe).

The tissues of the human body have been divided into the gelatigenous and the albuminous. Appropriate as such an arrangement might at first sight appear, it is opposed by the experience both of chemists and anatomists; Berzelius and E. H. Weber assert that as the permanent cartilages are not converted by boiling with gelatin, and as moreover they cannot be regarded as albuminous, cartilages must be divided into the gelatigenous and non-gelatigenous, and thus these observers abandon the old division of the tissues. Müller has subsequently devoted much attention to the structure and constitution of cartilage, and he finds that the permanent and fibrous cartilages which were previously regarded as non-gelatigenous, may be converted by very prolonged boiling into a gelatinizing and gluing substance; but at the same time he ascertained that in many of its other properties, this substance did not coincide with ordinary gelatin; hence he named it *cartilage-gelatin*, or *chondrin*.

Bone-gelatin or glutin is obtained from the following tissues, by boiling them for a longer or shorter time with water; from the cartilages of bone (after ossification), from tendons, the skin, calves' feet, hartshorn, isinglass, the scales of fish, and from the permanent cartilages, when they become ossified by disease. The conversion of these animal parts into glutin proceeds without any development of gas or absorption of air; acids promote this metamorphosis, just as they facilitate many similar transformations in organic chemistry, which can take place by mere boiling without their co-operation, but yet are hastened by their presence, as, for instance, in the case of starch.

We shall revert to this subject when treating of the individual tissues, and of their relation to gelatin.

*Origin.*—We have already referred to the production of gelatin from the gelatigenous tissues; a comparison of the analyses of pure gelatin with those of the tissues yielding it, will (in a future part of the work) show us that there is no chemical difference between the two, or that at most they only differ by a few atoms of water. Hence it appears that in the formation of gelatin, the material of the tissues only undergoes a rearrangement of its atoms, or a metamerism, or at most that it only assimilates water, just as occurs when starch, inulin, and lichenin are converted by prolonged boiling into dextrin or glucose.

We shall have occasion to refer in considerable detail to the production of gelatigenous from albuminous matters, when we treat of cell-formation and the history of development.

*Uses.*—From what has been already said, it follows that we are unable at present to discuss the uses of gelatin in the animal body.

<sup>1</sup> Arch. f. pathol. Anat. Bd. 5, S. 174.

The consideration of the tissues from which we obtain gelatin by boiling, pertains solely to histology, and the tissues themselves have as yet hardly fallen within the scope of chemical investigation. We learn from a very superficial consideration of the animal body that the gelatigenous tissues belong for the most part to the lower class of tissues, which are only of use through their physical properties; they frequently afford strong points of attachment for muscles, and furnish strong investments for important but easily injured organs; they give uniformity to the movements of the body through their elasticity, and protect it from the injurious effects of severe concussions; from being bad conductors of heat, they guard the body against rapid changes of temperature; and sometimes, as in the cornea, they are useful as refracting media, in consequence of their transparency.

### CHONDRIN.

#### *Chemical Relations.*

*Properties.*—Chondrin or cartilage-gelatin, when dry, appears as a transparent, horny, glistening mass, which is generally more colorless than glutin; it is not rendered electric by friction; its behavior towards indifferent solvents, towards heat, corrosive sublimate, tannic acid, and chlorine, is precisely the same as that of glutin; but its relations to acids and most metallic salts are quite different. It was shown by Müller<sup>1</sup> that *acetic acid* throws down a considerable precipitate from a solution of chondrin, and that this precipitate does not dissolve even in concentrated acetic acid. Simon<sup>2</sup> and Vogel<sup>3</sup> have subsequently proved that most acids throw down a precipitate from a solution of chondrin, but that this precipitate easily escapes notice in consequence of the facility with which it dissolves in a slight excess of the acid. *Alum, the sulphates of the protoxide and peroxide of iron, sulphate of copper, neutral and basic acetate of lead, and the nitrates of silver, and of suboxide of mercury* throw down copious precipitates. The precipitates thrown down by the salts of alumina occur in white, compact flocks, which on drying, cake very much together; they are insoluble in water, but dissolve in an excess of the earthy salt, as well as in solutions of chloride of sodium and of alkaline acetates. The precipitate thrown down by sulphate of peroxide of iron is not soluble in an excess of that salt, but dissolves on boiling. In its relations towards ordinary atmospheric influences, as well as towards alcohol, creosote, chlorine, bromine, iodine, and ferrocyanide of potassium, chondrin perfectly resembles glutin. Its combinations with other bodies and its products of decomposition have not yet been accurately studied.

Chondrin, when treated with sulphuric acid, yields, according to Hoppe,<sup>4</sup> no glycine, but only leucine. If sulphurous acid be passed through a warm solution of chondrin, the latter is at first precipitated, but afterwards undergoes decomposition with a development of ammonia

<sup>1</sup> Pogg. Ann. Bd. 38, S. 295.

<sup>3</sup> Journ. f. pr. Ch. Bd. 21, S. 426.

<sup>2</sup> Medicin. Chemie. Bd. 1, S. 108.

<sup>4</sup> Ibid. Bd. 56, S. 129.

and the formation of leucine and other products. On boiling with alkalis, chondrin is gradually decomposed with a development of ammonia. On treating it with a stronger solution of potash, or on fusing it with hydrated potash, there are formed glycine, leucine, and other products of decomposition. (Hoppe, however, could not find tyrosine.) In the putrefaction of chondrin there are formed, according to Hoppe, leucine and another crystallizable substance, in addition to other products of decomposition. On oxidation with chromic acid, it develops much prussic acid, but neither formic nor acetic acid.

Hoppe, who has more carefully analyzed chondrin than any of his predecessors, found 6.28% of salts in the substance in its ordinary state, and only 0.68% in chondrin treated with acetic acid.

The following is his method of preparing this substance: Cartilages are boiled for a short time, so as to effect the partial solution of the perichondrium, and, after its removal, they are cut into thin slices, macerated for some hours in cold water, and then boiled in a modified Papin's digester for 45 minutes or an hour, under a pressure of two or three atmospheres, by which means the greatest part of the cartilaginous substance is dissolved. On cooling the digester to 100°, the fluid is filtered as rapidly as possible, the filtrate evaporated, treated with cold water, the residue again dried, pulverized, extracted with boiling alcohol, and then dried at 120°. To remove the inorganic salts we must precipitate the solution of chondrin immediately after its first filtration with acetic acid, and after decanting the supernatant fluid, we must treat the precipitate with water; after the removal of the salts, it is, however, somewhat difficult of solution in boiling water.

*Composition.*—Mulder<sup>1</sup> was the first who made an elementary analysis of chondrin; he found that besides the ordinary elements of animal substances it contains a little free sulphur, and that it yields more than 4% of an ash consisting chiefly of bone-earth. It has subsequently also been analyzed by Scherer<sup>2</sup> and Schröder.<sup>3</sup> The following are the results of their analyses:

	Mulder.	Scherer.	Schröder.
Carbon, . . . . .	49.97	50.754	49.88
Hydrogen, . . . . .	6.63	6.904	6.61
Nitrogen, . . . . .	14.44	14.692	—
Oxygen, . . . . .	28.59	27.650	—
Sulphur, . . . . .	0.38		
	100.00	100.000	

From these results Mulder constructs the formula  $C_{32}H_{26}N_4O_{14}$ , and Scherer  $C_{43}H_{40}N_6O_{20}$ .

*Preparation.*—Chondrin is most readily obtained by boiling the cartilages of the ribs, larynx, or joints, for from 18 to 24 hours in water; to purify it we must adopt the same means as are recommended for gluten, and we must extract the dried residue with alcohol.

#### *Physiological Relations.*

*Occurrence.*—The remarks which have been already made regarding

<sup>1</sup> *Natuur en Scheik. Arch.* 1837, p. 450, and 1838, p. 160.

<sup>2</sup> *Ann. d. Ch. u. Pharm. Bd.* 40, S. 40–51.

<sup>3</sup> *Ibid.* Bd. 45, S. 52–58.

the occurrence of glutin in the animal organism, are equally applicable in relation to chondrin. Chondrin does not occur ready formed in the organism, but is produced by the prolonged boiling of certain tissues in water; all permanent cartilages in a healthy state yield chondrin on boiling. Müller's discovery that bone-cartilage not only yields chondrin before ossification, but also sometimes after it has undergone morbid changes, is very remarkable, and shows that chondrin and glutin, notwithstanding their perfectly different constitution, stand in a definite relation to one another; but what that relation is, we cannot at present conjecture.

There are, further, in the animal organism, several bodies which yield a gelatin distinct both from chondrin and glutin. Thus, Müller has shown that in *osteomalacia*, where there is sometimes a considerable diminution of the phosphate of lime, the bones yield neither glutin nor chondrin; that the *elastic tissue* of the arteries, by prolonged boiling, yields a kind of gelatin which only differs from chondrin in yielding no precipitate with sulphate of peroxide of iron; that the bones of cartilaginous fishes are converted by boiling into a substance which does not gelatinize but which glues very well, and which, moreover, resembles chondrin in its behavior to acetic acid and metallic salts, but is not precipitated by the salts of the oxides of platinum, silver, and gold; and, finally, that ossified fish-cartilage when boiled, yields a non-gelatinizing fluid, which is precipitated by tannic acid, but not by acetic acid and the salts of alumina, and consequently, approximates in its character to glutin.

*Origin.*—In our observations on glutin we pointed out that we are still perfectly ignorant of the mode of origin of chondrin. The experiments of Müller render it highly probable that glutin is formed from chondrin. But how? This must be decided by future researches.

*Uses.*—The animal tissues which yield chondrin are of the same use through their physical properties as those which yield glutin; their most important character being their elasticity.

## FIBROIN.

### *Chemical Relations.*

*Properties.*—It is a white, amorphous mass, devoid of odor or taste, insoluble in water, alcohol, and ether, but dissolving in concentrated sulphuric, nitric, and hydrochloric acids, from which solutions, if diluted with water, it is precipitated by tannic acid; it is insoluble in acetic acid and in ammonia; it dissolves in a concentrated solution of potash, but at the same time undergoes decomposition. This substance becomes decomposed, when heated; developing ammonia and empyreumatic vapors.

*Composition.*—This body was discovered and has been analyzed by Mulder;<sup>1</sup> it consists (taking the mean of four of his analyses) of:

<sup>1</sup> *Natuur en Scheik. Archief.* D. 3, p. 93, D. 5, p. 281.

Carbon, .	.	.	.	.	.	.	.	.	.	48·61
Hydrogen,	.	.	.	.	.	.	.	.	.	6·50
Nitrogen,	.	.	.	.	.	.	.	.	.	17·34
Oxygen, .	.	.	.	.	.	.	.	.	.	27·55
										<hr/> 100·00

From these numbers Mulder calculated the formula  $C_{39}H_{31}N_6O_{17}$ , according to which fibroin may be regarded as 3 atoms of glutin which have assimilated 1 atom of oxygen and 1 atom of water, for  $3(C_{13}H_{10}N_2O_5) + HO + O = C_{39}H_{31}N_6O_{17}$ . Mulder and Croockewit<sup>1</sup> moreover found that the common sponge contains the same substance in combination with iodine, sulphur, and phosphorus; and Mulder considers from the analyses of Croockewit that the compound consists of 20 atoms of fibroin, 1 atom of iodine, 3 atoms of sulphur, and 5 atoms of phosphorus; for there were found in sponge 1·08% of iodine, 0·50% of sulphur, and 1·90% of phosphorus, besides the elements of fibroin.

*Preparation.*—Silk or gossamer threads are boiled with water and strong acetic acid till all albuminous and gelatinous matters are dissolved. The remaining fibroin is then purified in the ordinary manner.

#### *Physiological Relations.*

This substance has hitherto been only found in the above-mentioned secretions of silk-worms and spiders; physiological investigations show us that it is originally a viscid fluid which is secreted by the spinning vessels of those animals, and hardens on exposure to the air. Under the microscope the fluid mass appears perfectly amorphous.

Sponge is, as is well known, the dry skeleton of an animal belonging to the *Porifera* (Grant) and named *Spongia officinalis* (Linn.) Its chemical constitution affords one of the arguments why the *Spongia* should be classed amongst animals and not amongst plants, since in the vegetable kingdom we nowhere meet with a substance in the slightest degree resembling fibroin.

The physiology of these lower animals has been so little investigated that it is impossible for us to set up an hypothesis regarding the formation of this substance, for notwithstanding the very accurate analyses of Mulder we cannot be regarded as knowing anything of its intimate chemical composition. Mulder's comparison of the composition of this body with that of gelatin, can indicate nothing more than the analogy in relation to the physiological value of both substances, that is to say, that nature produces in these lower animals a similar group of atoms in order to construct their solid groundwork of tissues possessing little or even no vitality. The use of this substance is therefore purely mechanical.

#### CHITIN.

##### *Chemical Relations.*

*Properties.*—This substance, to which Lassaigne gave the name of *Entomaderm*, is a white, amorphous body, which usually retains the form of the tissue from which it is prepared; it is insoluble in water, acetic acid, and alkalis, but dissolves in concentrated nitric and hydro-

<sup>1</sup> Scheik. Onderz. D. 2, p. 1.

chloric acids without communicating any color to those fluids; after neutralization with ammonia, tannic acid throws down a precipitate from these solutions. In concentrated sulphuric acid it swells up and becomes dissolved without communicating any change of color to the acid; it gradually however again separates as a black mass, while acetic acid and acetate of ammonia remain in solution; no sulphurous or formic acid is however formed. It is not decomposed by the most concentrated solution of potash, even at a boiling heat; heated to  $280^{\circ}$  with water in closed tubes, it becomes brown and brittle without undergoing any change of structure that can be detected by the microscope. There are two points worthy of notice in connection with the dry distillation of this substance; it does not fuse, but leaves a charcoal which on microscopic investigation always exhibits the form of the original tissue; and further, notwithstanding that it contains nitrogen, it yields acid products of distillation in which not only water and acetic acid are found, but also acetate of ammonia and a little empyreumatic oil.

*Composition.*—This body has been analyzed by Lassaigne<sup>1</sup> and Payen,<sup>2</sup> and has been most carefully studied by C. Schmidt.<sup>3</sup> Payen found much too little nitrogen. The results of various analyses and experiments which I have made with chitin exactly correspond with those of Schmidt. The following are the results of our analyses:

	Schmidt.	Lehmann. <sup>4</sup>
Carbon, . . . . .	46.64	46.734
Hydrogen, . . . . .	6.60	6.594
Nitrogen, . . . . .	6.56	6.493
Oxygen, . . . . .	40.20	40.179
	<hr/> 100.00	<hr/> 100.000

Schmidt regards  $C_{17}H_{14}NO_{11}$  as the simplest formula expressing this composition. He directs especial attention to the peculiar relations of this substance when acted upon by heat and by acids, and arrives at the very interesting result that this body, which so closely resembles vegetable bodies and especially vegetable fibre, may be regarded as composed of a carbo-hydrate similar to cellulose, and of a nitrogenous body which has the composition of the muscular fibre of insects. The latter is represented, according to his analyses, by the formula  $C_8H_6NO_3$ ; and  $C_{17}H_{14}NO_{11} - C_8H_6NO_3 = C_9H_8O_8$ .

*Preparation.*—The best method of obtaining this body is by boiling the elytra of the cockchafer with water, alcohol, ether, acetic acid, and alkalis; the body always perfectly retains the structure of the elytrum, or of the other insect-tissues from which it is prepared.

### *Physiological Relations.*

This body forms the true skeleton of all insects and crustacea. It constitutes not merely their external skeleton, the scales, hairs, &c., but also forms their tracheæ, and thus penetrates into the minuter portions of the organs; indeed even one of the layers of the intestinal

<sup>1</sup> Journ. de Chim. Méd. T. 9, p. 379.

<sup>2</sup> Compt. rend. T. 17, p. 227.

<sup>3</sup> Zur. vergleichend. Physiol. der wirbellos. Thiere, 1845, S. 32–69 [or Taylor's Scientific Memoirs, vol. 5, pp. 14–28.—G. E. D.]

<sup>4</sup> Jahresber. d. ges. Med. 1844, S. 7.

canal of insects consists of chitin ; hence we can very well prepare all these parts by treating insects with a solution of potash and then microscopically examine the finest parts, as for instance, the valves of the tracheal openings.

If Schmidt's hypothesis regarding the constitution of chitin be confirmed by further observations, it would be easy to understand how this substance is formed from the food of insects.

In reference to its application in the insect organism, chitin is at most entitled to be regarded as a histogenetic substance.

Before concluding our remarks on the organic substrata of the animal organism we would briefly review the mode of arrangement in which these substances have been considered. We observed in our remarks introductory to the subject of Zoo-Chemistry that the physiological and chemical classifications of animal substances must perfectly coincide with one another ; and now on our concluding observations we are constrained to admit that our knowledge of the organic substrata of the animal body is still very deficient, and that we have been provisionally compelled to adopt a practical classification and arrangement, in which, passing from the simpler to the more complex bodies, we have attempted to group together substances presenting chemical similarities with those of equal physiological importance. The deficiency of our knowledge on many points to which allusion has frequently been made, must plead as an apology for the deficiencies in our mode of arrangement. The laborious accumulation of properties, which are only slightly connected or are even altogether inapplicable, has grievously oppressed the science of chemistry, and has reduced it to a mere task of the memory. We have as yet no logical ideas in relation to chemistry ; that is to say, although we have perfectly clear perceptions regarding most bodies and processes, we have no distinct ideas (in the logical sense). There is an utter absence of those principles of unity around which, as around a nucleus, the individual properties of bodies can crystallize, and thus stand in the same mathematical relation to one another, as the edges and angles of crystal.

It is not till chemistry shall have shown us the close mutual connection that exist between the properties of all individual substances, and shall have taught us to unite them into one organic whole, that we can regard it as coequal in scientific rank with the different branches of physics,—that it will fully admit of the application of the higher mathematics,—or that the sole rational principle of classification as well as a scientific theory of chemical substances will be discovered. The beautiful investigations of Kolbe and others regarding the numerical ratio existing between the densities and boiling-points of the haloid bases, the volatile acids, and the haloid salts, as also the comparisons of the coefficients of density of the constituent elements with the other properties of the compound substance, may form a small beginning towards the attainment of logical ideas and the realization of such a degree of chemical knowledge. When we have once attained logical ideas regarding the different animal substrata,—when we are in a position to foretell the chemical properties of a body from its composition, or its composition from a certain number of its properties,—we shall then not only possess the true principle of

classification in physiological chemistry, but we shall also have attained the means of investigating and comprehending the vital processes of nutrition and secretion with a degree of certainty at present limited to the most exact sciences.

## MINERAL CONSTITUENTS OF THE ANIMAL BODY.

The chemistry of inorganic bodies has been so much more fully investigated than that of organic substances, that it might naturally be expected that our knowledge of the mineral constituents of vegetable and animal bodies would far exceed that of the organic constituents; but in truth, the reverse is the case, for we are far less acquainted with these substances than with many organic bodies. This circumstance is, however, not consequent on our having paid less attention to the mineral constituents of organic bodies, but it is especially owing to the difficulty of separating these substances, in an unchanged state, from organic matters, and of ascertaining the conditions and combinations in which they actually existed preformed in the organic substance. The fixed products of the incineration or combustion of organic substances do not afford us any information as to the combinations in which they occurred in the organic substance. Nor can any reflecting chemist for a moment suppose that the oxides and salts of the ash are contained as such in the juices and tissues of living bodies.

From a deficiency in the means of investigating or even of conjecturing the true constitution of these substances in organic parts, a higher value has been attached to the determinations of the ash and its constituents than it merited, and the results of these analyses have been more highly estimated than they deserve, when we consider the agents co-operating in the incineration. It has, moreover, frequently been forgotten that the quantity and constitution of many of the constituents of the ash are in a great measure dependent on the height of the temperature at which the process of incineration was conducted; that a great portion of the substances has been volatilized by the simultaneous action of heat and carbon; and that the individual constituents of the ash have entered into perfectly different combinations from what they had done in the organic substance.

We will here indicate only some few of the changes which the mineral constituents of organic substances must necessarily undergo when exposed to strong heat with a free admission of air. The sulphur and phosphorus which were not contained in the organic substance as sulphuric and phosphoric acids, must necessarily be found in the ash as sulphuric and phosphoric acids combined with bases; and although this necessary change has not been overlooked, the consequences have too often been neglected. When in the first place we direct our attention to sulphuric acid, we shall find that the number representing this acid as found in the ash, can scarcely ever correctly express the quantity of sulphuric acid existing preformed in the organic substance, or the sulphur contained in it. For if we suppose all the sulphur converted by combustion into sul-



phuric acid, and united to the bases that had previously been combined with organic substances or with carbonic acid, a great portion of the sulphur must be lost, even when these bases are sufficient for the saturation of the sulphuric acid that is formed (which is not always the case, as, for instance, in the bile) in consequence of the sulphates in contact with the nitrogenous charcoal, which is so difficult of incineration, being converted into metallic sulphides, of which a larger or a smaller quantity will escape as sulphurous acid during the prolonged process of calcination. Under the action of a strong glowing heat common phosphate of soda removes a part of the base, not only from the carbonates (see p. 97), but also from sulphates of the alkalis, as well as from the metallic chlorides of the ash, so that not only does all the alkaline carbonate disappear from the ash, but a portion of the hydrochloric or sulphuric acid may be also lost. Where the ash contains acid phosphate of soda, as occasionally happens in urine devoid of lactic acid, a portion of the phosphoric acid must necessarily be lost; for we know with what difficulty carbon burns in the presence of fusible salts, and it must be recollected that a portion of the phosphoric acid of the acid salts will be reduced by the carbon and volatilized. These few remarks may suffice to show how little attention was formerly directed to the reciprocal decompositions experienced by the mineral salts that occur in vegetable or animal substances, under the influence partly of a simple glowing heat, partly of heat in the presence of unconsumed carbon, and partly of a glowing heat in oxygen gas.

I have endeavored in some degree to evade these obstacles in the way of the determination of the mineral constituents of animal bodies, by isolating organic substances as much as possible, according to their solubility (as I have done in the case of blood,<sup>1</sup> for instance), and then determining the constituents of the ash of each separate extract; by which means we may be justified in expecting that the soluble salts that are preformed in the blood will be contained in the aqueous and alcoholic extracts, and that the presence of organic substances owing to their inconsiderable quantity in these extracts, will exert less influence on the decomposition of the salts during incineration. In order as much as possible to avoid the influence of the carbon and of the phosphates, during the process of incineration, on the carbonates, I have been in the habit of not exposing the whole of the carbonaceous residue originally obtained from the organic substance to entire combustion, but of reducing it to a small bulk over a gentle fire with free access of air. The carbonaceous ash is then extracted with water and hydrochloric acid, and the quantitative determination of the ash is obtained by weighing and subtracting the residuary charcoal. But although I have certainly obtained more correct results by this method than those yielded by the majority of previous analyses of ash, it is nevertheless not free from error, nor can it be said to afford an entirely satisfactory insight into the nature of the mineral substances existing preformed in animal bodies. Fortunately for science, H. Rose,<sup>2</sup> one of the most distinguished ana-

<sup>1</sup> *Berichte der k. sachs. Gesellsch. d. Wiss.* Bd. 1, S. 98.

<sup>2</sup> *Pogg. Ann.* Bd. 70, S. 449-465, *Berichte der Akad. der Wiss zu Berlin*, Decbr. 1848, S. 445-462, and *Pogg. Ann.* Bd. 76, S. 305-404. [The last of these memoirs is trans-

lysts of our day, has entered upon this hitherto unpromising subject, and by a series of the most carefully conducted investigations has obtained important results, which are in part of a purely physiological character. One of the most important facts ascertained by these successful researches in analytical chemistry is, that in the animal or vegetable substance perfectly carbonized by heat, there is usually a greater or lesser quantity of alkaline and earthy salts, which cannot be removed from the carbonaceous mass, even by the most prolonged extraction either with water or acids. These mineral substances must therefore be contained in the carbonized residue in a different condition from those which admit of being removed by various menstrua. Rose, therefore, concludes that such substances as alkalies, earths, metals, phosphorus, sulphur, &c., must be contained in the carbonaceous mass in a non-oxidized state, and in combinations with which we are still unacquainted: he also thinks that it may be assumed that such combinations of potassium, sodium, calcium, iron, phosphorus, and sulphur, also exist preformed in organic substances, since on the one hand the carbonization of organic substances free from ash (as for instance sugar) with the ordinary constituents of the ash did not yield any carbonaceous residue that could not be perfectly freed by the ordinary menstrua from mineral substances; and since, on the other hand, we are already acquainted with some organic bodies in which we assume that non-oxidized sulphur or non-oxidized iron is present in a peculiar state of combination. Hence Rose further concludes that in vegetable and animal substances those mineral constituents can alone be regarded as preformed, which admit of being extracted by means of water and acids from the carbonized material, while on the other hand those substances which cannot be separated until the carbonaceous mass is entirely burned, are inherent in the original organic substance, as integral constituents in a non-oxidized condition.

It appears from the numerous investigations prosecuted by Rose, with vegetable and animal products, that while there are some, as, for instance, the bones, in which all the mineral constituents are in a perfectly oxidized state, that is to say, admit of extraction by the ordinary solvents (and these he names *teleoxidic* organic substances), the great majority contain the mineral constituents partly in an oxidized and partly in an unoxidized state (these he terms *meroxidic*), while none are as yet known that contain only unoxidized elements (*anoxidic*).

In his examination of vegetable substances, Rose found that the straw of different kinds of grain was almost perfectly *teleoxidic*, whilst the seeds of the same plants were *meroxidic*. In reference to animal substances, it was to be expected that, as the *meroxidic* substances belonging to the vegetable kingdom specially serve as food for the animal organism, those animal fluids and tissues whose chemical constitution approximates to that of vegetable substances, as the blood, the muscular fibre, milk, and yolk of egg, would be *meroxidic*, whilst the excretions, as matters which originated in the animal body mainly by the process of oxidation, would be *teleoxidic*. This supposition has been fully confirmed by the

analyses of the bile, the urine, and solid excrements, instituted by Weber, Fleitmann, Weidenbusch, and Poleck. In order to take a general view of these relations, we will subjoin the numerical results which have been obtained, according to Rose's method, by investigations on the mineral constituents of animal substances. In the following table, A represents the quantity of the salts that can be extracted by water from 100 parts of the mineral constituents of the organic substance; while B represents the quantity of salts dissolved by hydrochloric acid; and C, the quantity of the salts which can only be determined by the combustion of the carbonaceous residuc.

	A.	B.	C.
Ox-blood, . . . . .	60.90	6.04	33.06
Horseflesh, . . . . .	42.81	17.48	39.71
Cows' milk, . . . . .	34.17	31.75	34.08
Yolk of egg, . . . . .	40.95	8.05	51.00
White of egg, . . . . .	82.19	15.52	2.29
Ox-bile, . . . . .	90.85	4.93	4.22
Urine, . . . . .	90.87	8.54	0.59
Solid excrements, . . . . .	18.55	62.30	19.15

The column C exhibits, therefore, those mineral substances in the oxidized state, which, according to Rose, are not oxidized in the organic substance.

It must be further observed, that in the solid excrements the number representing the mineral substances that cannot be extracted, would not be so strikingly high if sand and the silica of the vegetable tissue were not mixed with them; the number representing the non-oxidized substances is also increased in the white of egg, the ox-bile, and the urine, by the silica occurring in them.

Although Rose's investigations have greatly contributed to our advance towards the knowledge of the inorganic constituents of animal substances, we dare not flatter ourselves that we have as yet attained the object in view, for it not only remains for us to apply this method to the investigation of the mineral substances contained in different normal and morbid animal juices and tissues, but also, by further investigation, definitely to determine the question that has been started against Rose's view of the combination of radicals containing sulphur and phosphorus with metals; in other words, it will be necessary to collect a greater number of facts, in order to illustrate this obscure subject in various points of view, before we venture to apply it, in all its consequences, to scientific questions. Yet it cannot be denied that no previous method affords us so good a guide as Rose's for the correct recognition of the mineral substances existing preformed in organic bodies.

When, however, we have obtained by Rose's method such an admixture of mineral bodies as we may assume to exist preformed in the organic substance, the actual analysis still remains to be made; and this, notwithstanding the labors of the most eminent chemists, has by no means attained to the degree of perfection which has been generally obtained in mineral analyses. The recent investigations of Fresenius, Erdmann, Mitscherlich, and more especially of Rose, have made us acquainted with numerous deficiencies which attached to the former methods of examining the ashes of vegetable and animal substances; and not-

withstanding this, we are struck with the great accuracy of many of the earlier analyses of ashes, although from the methods then employed we should have expected that their calculations would of necessity have yielded a *minus* in the one case, and a *plus* in the other.

We will here only refer to the fact that few observers before Rose had observed that alkaline as well as earthy salts were contained in the insoluble portion of the ash, and that, conversely, the presence of carbonate and phosphate of lime in the aqueous extract of the ash had been very generally overlooked, while the very imperfect precipitation of the pyrophosphate of magnesia by ammonia was equally disregarded. The imperfect manner in which even the simplest relations of this nature have been investigated, is made apparent by the doubts entertained by Berzelius himself, in reference to the composition he had ascribed to bone-earth, which were verified by the investigations of Rose and W. Heintz,<sup>1</sup> by whom it was definitely proved that the phosphate of lime in the bones is represented by  $3\text{CaO} \cdot \text{PO}_5$ , and not as Berzelius had given it, by  $8\text{CaO} \cdot 3\text{PO}_5$ . The difficulty of conducting exact analyses of ash was, however, mainly increased by the deficiency of any clear and comparatively simple method of separating phosphoric acid from its proteus-like salts, and determining it quantitatively. But this cause of difficulty has likewise been recently obviated by H. Rose's<sup>2</sup> method of thoroughly separating the acids from their bases by means of mercury and nitric acid.

When we consider these facts in reference to the analysis of the ash, we shall readily arrive at the conclusion (without, however, wishing to animadvert upon those analysts who have engaged in laborious examinations of the ash of animal bodies), that most of these analyses should be used with great caution, and that physiological conclusions should not be too readily drawn from them. It has, unfortunately, too often happened that the empirical results of analyses of the ash have been applied to the explanation of physiological processes without due consideration, and thus the importance and efficiency of the mineral salts of the animal body have been extolled before we had any accurate knowledge of the substances themselves; and the most rigorous scepticism in reference to medical experiments has not unfrequently been associated with a blind confidence in the least reliable of the numerical determinations of chemists.

Since we have made a practice of incorporating the methods of qualitative and quantitative analysis in the description of the organic substrata, it might naturally be expected that we should in like manner enter into a special consideration of the different methods for analyzing the ash; but however important this subject may be, both in itself and in reference to physiology, we have, nevertheless, been deterred by many reasons from adhering to this rule in the present case. Thus, for instance, if we were once to enter thoroughly within the domain of inorganic chemistry, we should far exceed the limits assigned to this work, more especially if we were definitely to refer to, and critically to illustrate, the different methods for the analysis of the ash and the determination of individual constituents; nor could we indicate any one method as the best, since different objects demand different methods.

<sup>1</sup> Ber. der Ak. d. Wiss. z. Berlin, Febr. 1849, S. 50-53.

<sup>2</sup> Ibid. S. 42-45.

We, moreover, entertain the frequently expressed, but rarely practised view, that the study of physiological as well as of organic chemistry, generally, should be based upon an exact knowledge of inorganic chemistry in all its relations, for many of the deficiencies which we have found occasion to notice in the researches of zealous physiological and pathological chemists, are referable to an inadequate knowledge of inorganic chemistry. We are, therefore, the more resolved to omit all notice of the analyses of mineral substances, again referring our readers to the admirable memoirs which have appeared in recent times on this subject, and for which we are indebted to Will and Fresenius,<sup>1</sup> Mitscherlich,<sup>2</sup> Knop,<sup>3</sup> Erdmann,<sup>4</sup> Heintz,<sup>5</sup> Rose<sup>6</sup> [and Strecker.<sup>7</sup>—G. E. D.]

If we venture to adopt a physiological classification in our description of the mineral substances of the animal body (which, moreover, can refer only to their physiological function), we adopt this course simply from a feeling of its great applicability, and not because we consider ourselves able to indicate the exact place occupied in this system by each individual mineral substance; for the remarks we have already made, must sufficiently indicate the uncertainty and deficiency of our knowledge on this subject. We therefore attempt to divide the mineral substances of the animal body in reference to their physiological importance, into :

1. Those which are of especial use in the animal body through their physical properties.

2. Those which are adapted by their chemical properties to serve definite objects in the animal economy: and

3. Those which are only incidentally conveyed into the animal body, exert no influence on any special process, and are, therefore, speedily eliminated from the organism.

#### FIRST CLASS OF MINERAL BODIES.

##### WATER.

It would be superfluous to enumerate the uses of this substance in the animal organism; we will confine ourselves to the two simple remarks that water is essential to the establishment of all chemical activity, and, further; that the functions, or rather the physical properties of certain tissues, are dependent on the presence of a certain quantity of water, which is merely in a state of mechanical combination.

#### PHOSPHATE OF LIME.

This is the most important of all the mineral substances which, by their physical properties, are of service in the animal body. The use of its presence in the bones, where it gives solidity and strength to the

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 50, S. 363-396.

<sup>2</sup> Ber. d. Akad. d. Wiss. z. Berlin, 1845, S. 236-252.

<sup>3</sup> Journ. f. pr. Ch. Bd. 38, S. 14-47.

<sup>4</sup> Ibid. Bd. 38, S. 40-69, and Ber. d. Gesellsch. d. Wiss. zu Leipzig, 1847, S. 83-90.

<sup>5</sup> Op. cit.

<sup>6</sup> Op. cit.

<sup>7</sup> Ann. d. Ch. u. Pharm. Bd. 73.

osseous skeleton, is at once apparent. Bones, deficient in this salt, are proportionally deficient in firmness: thus we observe that softening of the bones occurs in those conditions when the animal organism does not receive a sufficient supply of phosphate of lime, or when certain physiological processes require an increased consumption of this salt, as in pregnancy, and during the dentition of children. We need hardly remark that rachitis frequently, if not always, occurs simultaneously with the period of dentition, that the consumption of phosphate of lime during pregnancy is often so great that scarcely any traces of it can be found in the urine, and that, during this period of woman's life fractures unite with extreme difficulty, and sometimes do not unite at all. Chossat<sup>1</sup> was able to induce softening of the bones artificially in animals, when he restricted them to food containing little or no phosphate of lime. The permanent cartilages only ossify in old age, when a superabundance of calcareous salts is deposited in them. In the dense, cortical portion of bones, we find more bone-earth deposited than in the spongy parts. The teeth, whose utility depends entirely on their hardness, contain a larger proportion of phosphate of lime than any other part of the animal body; and it exists in still greater quantity in the enamel than in the dentine.

We have previously had occasion to remark that Berzelius, even to a recent time, adhered to the formula  $8\text{CaO} \cdot 3\text{PO}_5$  for the phosphate of lime of bone-earth, and that on the other hand, the investigations of W. Heintz under Rose's direction, indicate that the formula for the composition of bone-earth should be  $3\text{CaO} \cdot \text{PO}_5$ . Berzelius<sup>2</sup> has in part given the reason for his formula. It is not always  $8\text{CaO} \cdot 3\text{PO}_5$ , which is precipitated from acid solutions containing lime and phosphoric acid, as he formerly assumed; but when there is an excess of lime, and under the prolonged action of caustic ammonia, the basic salt  $3\text{CaO} \cdot \text{PO}_5$  is precipitated. Since the phosphate of lime is for the most part separated in this way, and the lime which is precipitated after the removal of the phosphate is calculated as if it were a carbonate, without any direct determination of the carbonic acid, there must be some uncertainty in the ordinary analyses of the earthy constituents of the bones, in part owing to the not very accurate determination of the magnesia. Heintz has found that this is the composition of phosphate of lime not only in normal human bones, but also in those of the sheep and the ox. In this point of view, however, the investigation of diseased bones requires a thorough revision; moreover, Von Bibra's<sup>3</sup> analyses seem to show that in the teeth, the ratio of the phosphoric acid to the lime is not in accordance with either of the above formulæ.

In healthy human bones the phosphate of lime ranges from 48 to 59%; in softening of the bones it may sink to 30%. It is, however, singular that in almost all diseases of the bones, whether the results of osteoporosis, osteomalacia, or osteopsathyrosis, we find a diminution of the phosphate of lime. Even in consecutive induration (or eburneation), the bones often do not regain their normal quantity of phosphate of lime.

<sup>1</sup> Gaz. méd. 1842, p. 208.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 286-289.

<sup>3</sup> Chem. Unters. üb. Knochen u. Zähne. Schweinfurt, 1844, S. 284-287.

Von Bibra has very fully investigated the composition of the different bones of the same individual, and has made the beautiful observation that those bones which are the most exposed to mechanical influences, contain the largest amount of earthy constituents. The action of this law is manifested even in different families of the same class of animals; thus, for instance, in the rases or scraping birds, the femur contains the largest quantity of phosphate of lime, in the grallatores or waders, the tibia, and in all other birds, the humerus.

That the phosphate of lime and the earths generally are only mechanically deposited in the bones, is obvious from the circumstance that we can so thoroughly deprive them of all mineral constituents by dilute hydrochloric acid, that they leave scarcely a trace of ash.

It has for a long time been a matter of discussion whether the phosphate of lime is, or is not, chiefly deposited in the bone-corpuscles and the *canaliculæ chalicophoræ*. I am, however, now convinced that the dark color of these parts in refracted light, and their white color in reflected light, essentially depends on their containing air. Any one may readily convince himself that this is the case, by treating one thin section of bone with dilute hydrochloric acid, so as to remove the earths, and another with a dilute solution of potash, so as to remove the cartilaginous substance, and comparing the two under the microscope. Frerichs<sup>1</sup> attempted to demonstrate that the earths were uniformly distributed throughout the bone by showing that osseous laminæ from which the cartilaginous substance had been removed by a dilute solution of potash, received a uniform yellow tint on the addition of nitrate of silver, and that the bone-corpuscles were not distinguished by any special depth of color.

Phosphate of lime also occurs in many other parts of the animal body, although in far less quantity than in the bones; indeed there is no animal tissue, in whose ash, or incineration, we do not find phosphate of lime.

Liebig<sup>2</sup> regards the insolubility of certain tissues, as for instance muscular fibre and cellular tissue, as partially due to the bone-earth which they contain. In the transition of the blood into these tissues its protein-compounds part with the soluble phosphate of soda, but retain a large quantity of the phosphate of lime. It is thus, that Liebig accounts for the special power which hydrochloric acid possesses of dissolving these substances during the process of digestion.

Well-dried muscular fibre contains, according to von Bibra, from 0.938 to 1.008% of bone-earth.

Phosphate of lime is found in solution in all the animal fluids; its presence has long been recognized in the blood, the urine, the fluids of serous membranes, the saliva, gastric juice, milk, and seminal fluid, but it was for a long time unknown by what means this insoluble body was retained in solution in alkaline and neutral fluids. As a general rule phosphate of lime is chemically combined with the protein-compounds and similar organic matters, and is retained by them in their solutions

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 43, S. 251.

<sup>2</sup> Ibid. Bd. 50, S. 170.

as well as in their metamorphoses into the tissues. Moreover it has been long demonstrated by Berzelius and Thenard, that phosphate of lime is to a certain degree soluble in fluids containing much carbonic acid; we know from analytical chemistry, that it is not altogether insoluble in fluids containing hydrochlorate of ammonia, and recently Liebig has shown that a little phosphate of lime is taken up by solutions of chloride of sodium. The solubility of bone-earth in animal fluids is thus sufficiently intelligible.

We have already spoken of the solvent power which lactic acid exerts on phosphate of lime. In opposition to the experiments of Walter Crum<sup>1</sup> I will only remark that in my experiments (taking the mean of six) 68·55 parts of basic phosphate of lime were dissolved by 100 parts of anhydrous lactic acid, while a fluid containing 100 parts of anhydrous acetic acid could only dissolve 17·49 parts of the same salt.

The ash of the protein-compounds consists for the most part of phosphate of lime; Berzelius<sup>2</sup> found 1·8% in the albumen from the serum of ox-blood, while Mulder found 2·03% and Marchand from 2·1 to 2·5% in that of the egg; in soluble albumen precipitated by great dilution and neutralization, I found 1·3% of phosphate of lime; in well-washed fibrin from the venous blood of a man, I found only 0·694%. Casein, globulin, chondrin, and gluten also contain phosphate of lime as an integral constituent. Casein, according to Mulder,<sup>3</sup> contains 6% of phosphate of lime, which, when the casein is coagulated, is precipitated with it, even when there is a sufficient quantity of free acid in the fluid. Chondrin, according to Mulder, yields on incineration 4·09% of ash, most of which is phosphate of lime. As chemical compounds of phosphate of lime with albumen and with gelatin have been prepared, which contain much greater quantities of the salt (in albumen even one-third) there would be nothing absurd in the supposition that a portion of the phosphate of lime contained in the bones, is chemically combined with the cartilaginous substance, even though it may be removed by hydrochloric acid.

The constant occurrence of phosphate of lime in the histogenetic substances, and especially in the plastic fluids, as well as its deposition in many pathologically degenerated cells of the animal body, obviously strengthen the opinion that this substance plays an important part in the metamorphosis of the animal tissues, and especially in the formation and in the subsequent changes of animal cells. This subject must, however, be more fully investigated, before we can draw any definite conclusions regarding it.

In connection with this subject, C. Schmidt<sup>4</sup> has, however, made a very interesting observation regarding the folds of the mantle of *Unio* and *Anodonta*. They consist of a middle layer of fibres of areolar tissue, which on its inner side is covered with ciliated epithelium and towards the shell with glandular epithelium; in these parts he found about 15% of phosphate of lime, 3% of carbonate of lime and soluble salts, and 82% of organic matter,—the quantity of phosphate of lime being very extraordinary, as the blood of these animals contains only 0·034% of this

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 63, S. 394 ff.  
Archiv. f. 1828, p. 155.

<sup>2</sup> Lehrb. d. Ch. Bd. 9, S. 35.

<sup>4</sup> Zur vergleichenden Physiol. S. 58–60.



salt. The mucus, lying between the shell and the mantle of these animals, and secreted by the layer of glandular cells on the mantle for the consolidation of the shell, consists of a strong basic albuminate of lime containing only a little preformed carbonate of lime. Schmidt is of opinion that the function of this glandular epithelium, which resembles the cells of the liver, is to secrete from the blood a combination of albumen and lime, decomposable by the carbonic acid of the air or of water, for the formation of the shell, while it leaves the phosphate of lime for those organs which require it for the process of cell-formation (the testicle and ovary).

The questions now arise, how do such masses of phosphate of lime find their way into the animal body? Or how are they formed in it? That carnivorous animals receive a more than sufficient quantity with their food is obvious from the preceding observations. Graminivorous animals likewise receive in their food a sufficient quantity of this earthy salt; for in the vegetable kingdom, we find certain nitrogenous bodies, which, like the protein-compounds of the animal organism, always contain some phosphate of lime, as for instance, vegetable albumen, legumin, and gluten.

Phosphate of lime, is, however, also formed within the animal organism. If the experiments of von Bibra, showing that the bones of young creatures contain relatively more phosphate of lime than those of older ones, appear to be opposed to the view that the phosphate of lime is formed from the carbonate, the numerous analyses of Valentin<sup>1</sup> prove that newly formed bones, or parts of bones, always contain a greater quantity of carbonate of lime before they are provided with their proper quantity of phosphate of lime. If we review the different substances taking part in the metamorphosis of the animal tissues, it appears, as a necessary conclusion, that phosphate of lime must be formed from its proximate constituents. We know that several animal substances contain phosphorus in an unoxidized state, and that they are not removed from the organism till they are perfectly decomposed, that is to say, till they are partially oxidized; in this process the phosphorus must be converted into phosphoric acid. We further know that very many animal substances also contain sulphur, and in their decomposition in the animal body form not only sulphuric acid, but also uric, hippuric, and other acids, which must partially decompose the alkaline phosphates that find their way into the body from without, that is to say, by the seeds of the cereals and leguminous plants, so that the liberated phosphoric acid must combine with the lime which enters the animal body with the vegetable food or with the water used as drink. We have an opportunity of almost directly observing the process of the new formation of phosphate of lime from its proximate constituents in the development of the chick within the egg; for the observations of Prout and Lassaigne show that during incubation, such a quantity of carbonate of lime is transferred from the shell of the egg to the yolk, that the augmentation of the phosphate of lime with the growth of the chick during incubation, is not more than can be accounted for.

Valentin's opinion is based on the following observations:—In the

<sup>1</sup> Repert. f. Anat. u. Physiol. 1839, S. 306 ff.

carious tibia of a man, aged 38 years, he found 44·12% of ash containing 77·93% of phosphate, and 15·04% of carbonate of lime, while the tibia of a healthy man of the same age yielded 61·98% of ash, in which were contained 84% of phosphate, and 12·8% of carbonate of lime. Hence, in this case the amount of ash was diminished almost solely at the expense of the phosphate of lime. In the callus, as well as in the exostosis of a horse, he found the carbonate of lime increased in relation to the phosphate, and hence concluded, that, as a general rule, imperfectly formed bones always contain more carbonate of lime than normal bones. Lassaigne's experiments<sup>1</sup> accord with those of Valentin. In the osteophyte occurring on the inner layer of the skull during pregnancy, there is also much carbonate of lime, as was observed by Kühn; I found 52·46% of organic matter, 30·69% of phosphate of lime, 1·09% of phosphates of magnesia and iron, 0·98% of soluble salts, and 14·78% of carbonate of lime in one of these osteophytes.

Prout<sup>2</sup> was the first who observed that during the incubation of the egg the quantity of phosphorus in its contents remains constant, but that the quantity of lime undergoes a considerable augmentation; he was almost inclined from this observation to conclude that there was a formation of lime from other materials, since he did not regard it as probable that the non-vascular *membrana putaminis* could transfer lime from the shell to the embryo. But if we take into consideration that during incubation the shell experiences a loss both in weight and firmness, and that a part of this *membrana putaminis* becomes dried, and consequently impermeable, while, however, the greater part is in contact with the contents and thus remains moist, it is very easy to perceive that the increase in the amount of lime within the egg arises from its most proximate source, namely, from the shell itself. The phosphorus exists chiefly in the yolk, where it occurs as glycerophosphoric acid, which during incubation is gradually decomposed, so that the liberated phosphoric acid unites with lime which passes over by endosmosis from the shell into the egg to form this salt. There is, however, so much phosphorus contained in the yolk of the egg, that on incineration it forms acid phosphates, or rather metaphosphates ( $\text{NaO.KO.PO}_5$ ), with the bases which it there encounters.

### CARBONATE OF LIME.

This salt is principally found in the skeletons of invertebrate animals; but it always occurs, as has been already mentioned, in greater or smaller quantities, in the bones of the vertebrata. Its uses in the animal organism are the same as those of phosphate of lime.

There can be no doubt that the carbonate of lime found in animal substances is very often no educt, but the product of the incineration to which we have submitted the substance in the course of the chemical analysis; it not unfrequently, however, occurs in the bones of the vertebrate animals as true carbonate of lime, and in the lower classes of this

<sup>1</sup> Journ. de Chim. Méd. T. 4, p. 366.

<sup>2</sup> Phil. Trans. 1822, p. 365.

great division we find it deposited in various places in microscopic crystals. Carbonate of lime in considerable quantity is found in the urine of graminivorous animals, in the saliva of the horse, and in many animal concretions.

Numerous experiments have been instituted, especially by Lassaigne, Fernandes de Barros,<sup>1</sup> Valentin,<sup>2</sup> and von Bibra,<sup>3</sup> with the view of ascertaining the ratio in which the carbonate of lime stands to the phosphate in the bones of different men and animals. According to my own investigations, this ratio in a new-born child = 1 : 3·8, in an adult male = 1 : 5·9, and in a man aged 63 years = 1 : 8·1; according to Valentin it = 1 : 8·3 (on an average) in caries, and = 1 : 5·54 in callus, or 1 : 5·3 according to Lassaigne; in an exostosis it = 1 : 52 according to Valentin, and 1 : 1·214 according to Lassaigne; according to Barros it = 1 : 3·8 in the lion, 1 : 4·15 in the sheep, 1 : 8·4 in the hen, 1 : 3·9 in the frog, and 1 : 1·7 in a fish. According to Lassaigne this ratio = 1 : 3·6 in the teeth of a new-born child, 1 : 5·3 in those of a child aged six years, 1 : 6 in those of an adult, and 1 : 6·6 in those of a man aged 81 years.

Von Bibra, in his numerous analyses of bone, has arrived at opposite results, since he found that the bones of young creatures for the most part contained less carbonate of lime than those of older ones. As we must refer for fuller information to von Bibra's work, we shall here only give the quantity of carbonate of lime which he found in the femur in different classes of animals; in the order *glires*, it amounts to 9·48%, in the *ruminantia* to 9·86%, in the *pachydermata* to 10·15%, in the *cetacea* (the dolphin) to 9·99%, in the *pinnipedia* (the seal) to 7·23%, in the *falculata* to 6·26%, in the *pollicata* to 9·18%, and in men to 8·59%.

The urine of graminivorous animals often contains so large a quantity of carbonate of lime as to cause a deposit very soon after its emission. My investigations tend to show that in the urine of the horse carbonate of potash and carbonate of lime very frequently replace one another; I have usually found that urine rendered turbid by the presence of much carbonate of lime contains a very small quantity of alkaline carbonates, and often has only a very slight reaction on turmeric paper, while clear urine is usually rich in alkaline carbonates. Hence it is easy to see why urinary calculi consisting of carbonate of lime are of very common occurrence in herbivorous animals.

Carbonate of lime sometimes also occurs in human urine with an alkaline reaction; and indeed sometimes, although very rarely, we meet with human urinary calculi, consisting for the most part of carbonate of lime. Prout<sup>4</sup> was the first who made this observation; but similar calculi have been since found by Cooper, Prout,<sup>5</sup> Smith, Gübel,<sup>6</sup> and Fromherz.<sup>7</sup>

In animal concretions, we sometimes find considerable quantities of carbonate of lime deposited with the phosphate. Thus, Geiger<sup>8</sup> found 21·7 of carbonate and 46·7 of phosphate of lime in a nasal concretion; I

<sup>1</sup> Journ. de Chim. méd. T. 4, p. 289.

<sup>2</sup> Op. cit.

<sup>3</sup> Thomson's Annals of Philos. vol. 15, p. 436.

<sup>4</sup> Trommsdorf's n. Journ. Bd. 9, S. 198.

<sup>5</sup> Mag. f. Pharm. Bd. 21, S. 247.

<sup>6</sup> Op. cit.

<sup>7</sup> A. Gehlen's Journ. Bd. 3, S. 582.

<sup>8</sup> Schweigg. Journ. Bd. 46, S. 329.

found 24·3% of carbonate and 69·7% of phosphate of lime in a phlebolith, and Schlossberger<sup>1</sup> 8·3 of carbonate and 50·4 of phosphate of lime in a similar concretion; Walchner<sup>2</sup> found 23% of carbonate and 50% of phosphate of lime in a concretion from the heart of a man with hydrothorax, and John<sup>3</sup> found 66·7% of carbonate and 25% of phosphate of lime in a concretion taken from a stag's heart. Some stony concretions, from the peritoncum of a man were found by Bley<sup>4</sup> to contain 34% of carbonate and only 19·32% of phosphate of lime; Lassaigne<sup>5</sup> found 83·36% of carbonate of lime in a salivary concretion from a horse. I need hardly advert to the frequency with which we meet with tolerably large quantities of carbonate of lime in the microscopico-chemical investigation of indurated or ossified tumors, as for instance, chalky tubercle.

Carbonate of lime in the crystalline state is very rarely found in the human organism; the only place where it constantly occurs in the normal state is the *utriculus* of the membranous vestibule<sup>6</sup> of the inner ear, on whose outer and upper walls it is deposited in minute crystals amongst organic matter. These crystals are usually so very minute, that distinct molecular motion may be observed amongst the smallest of them. The form of the crystals is never a pure rhombohedron, but always a prism derivable from the rhombohedron of calc-spar, most frequently resembling the so-called *Kanonendrusen* of calc-spar;<sup>7</sup> that is to say, they are six-sided with 3-planed acuminations. Krieger<sup>8</sup> has also seen twin crystals of the scaleno-octahedral form. Crystals of this nature occur much more frequently and abundantly in the lower animals, both in the organs of hearing and in other parts; perhaps the best known and most striking case of the occurrence of such crystals is in the membrane of the brain of the *batrachia*, and in the white, silvery saccules at the intervertebral foramina through which the spinal nerves emerge. In morbid formations in the human organism, we not unfrequently meet with crystalline deposits of carbonate of lime, which however usually appears rather in irregular crystalline masses, such as are described by Vogel,<sup>9</sup> than as perfectly formed crystals.

There are obviously two ways in which we may account for the presence of carbonate of lime in the animal organism. It is well known that spring water holding carbonic acid in solution, usually contains a considerable quantity of carbonate of lime; and this might sufficiently explain the presence of this salt, even if it were not in a great measure formed within the organism from other salts of lime, which find their way there in abundant quantity with the vegetable articles of food; hence it is that the urine of herbivorous animals is often so rich in carbonate of lime.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 69, S. 254.

<sup>2</sup> Mag. f. Pharm. Bd. 19, S. 152.

<sup>4</sup> Arch. d. Pharm. Bd. 20, S. 212.

<sup>3</sup> Chem. Schriften. Bd. 5, S. 155.

<sup>5</sup> Journ. de Chim. Méd. 1845, p. 523.

<sup>6</sup> [It occurs also in the *sacculus*, and is sometimes scattered in the cells lining the *ampullæ* and semicircular canals.—G. E. D.]

<sup>7</sup> [The term *Kanonendrusen* is used in the Hartz to signify a crystalline modification of calc-spar. *Drusen* signifies a cluster of crystalline substances. A crystal is said to be *drusy* (*drusig*) when it is coated with a number of minute crystals of the same kind, so that the new surface acquires a scaly aspect.—G. E. D.]

<sup>8</sup> De otolithis, Berolini, 1840, p. 15.

<sup>9</sup> Icones histol. path. Tab. 22, fig. 8.

The solubility of this salt in the animal fluids, might, at first sight, seem to be less easily understood than its origin. The free carbonic acid which, it is almost certain, may be detected in all the animal fluids, doubtless acts as a solvent for the carbonate of lime; and I may remind any who may not be satisfied with this explanation, that the old experiments of Guiton Morveau, show that carbonate of lime is also slightly soluble in solutions of the alkaline salts, as for instance, chloride of potassium. Moreover, it is not improbable that there are several animal substances which, like sugar, exert solvent action on carbonate of lime.

### PHOSPHATE OF MAGNESIA.

Phosphate of magnesia always occurs in such small quantity that we feel scarcely justified in ascribing to it simply a mechanical use in the animal body, and in arranging it in this class of the mineral substances; it is, however, so constantly associated with the corresponding lime-salt that we feel compelled to notice it in this place. Like the phosphate of lime, it is in the osseous system that it is chiefly deposited.

The bones of carnivorous animals and of man contain very little phosphate of magnesia; those of herbivorous animals rather a larger quantity. Berzelius found 1.16% in a piece of human bone, and 2.05% in the bones of an ox; Valentin found 1.943% in a portion of one of the ribs of a horse; Berzelius 1.5% in the enamel of a human tooth, and 3% in that of the tooth of an ox; in human dentine he found 1%, and in that of the ox 2.07%. The numerous analyses of von Bibra afford a general confirmation of these facts; he observed, moreover, that the teeth of the *pachydermata* were especially rich in phosphate of magnesia. Various physiological relations (age, &c.), as well as morbid conditions, augment and diminish the quantity of this salt, which seems, however, to vary in a direct ratio with the phosphate of lime. We shall return to this subject in our remarks on "The Bones."

That a little phosphate of magnesia occurs in all the animal fluids and tissues is demonstrated by the analyses of the ash. The presence of this salt is very strikingly shown by a microscopic examination of the tissues of a dead body in which putrefaction has actively commenced: we observe that it is everywhere studded with the well-known crystals of the phosphate of ammonia and magnesia.

Phosphate of magnesia sometimes accumulates in large quantities in certain concretions; thus Brugnatelli<sup>1</sup> found a concretion in a human ovary consisting almost entirely of this earthy salt, and a similar one in the uterus, which was surrounded by a thin crust of phosphate of lime. A phlebolith, examined by Schlossberger<sup>2</sup> contained 58.7% of salts of lime, 13.7% of phosphate of magnesia, and 20.4% of organic matters.

The *origin* of the phosphate of magnesia is sufficiently obvious; for this salt occurs in all parts of plants, and particularly in the common varieties of grain that are used for food. From the ratio in which, as we have shown, the phosphate of magnesia stands to the phosphate of lime in the bones and other parts, we may conclude that the animal

<sup>1</sup> Brugn. Giorn. T. 12, p. 164.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 69, S. 254.

economy requires far less of this salt than of the corresponding lime-salt; and this is especially illustrated by the fact that in different animals it is found that the intestinal canal absorbs all the phosphate of lime, but only very little phosphate of magnesia; for the excrements of the *carnivora*, as well as of the *herbivora*, contain an excess of the latter salt.

From these facts, Berzelius' long ago drew the conclusion that the absorbents of the intestinal canal have less tendency to take up phosphate of magnesia than phosphate of lime, but that rather more is always absorbed by the *herbivora* than by the *carnivora*; this latter fact, however, probably depends upon the circumstance that the food of the former contains far more magnesia than that of the latter class of animals. We should, however, be too strictly interpreting the meaning of Berzelius if we were to suppose that he considered the absorbents to possess any special power of selecting and taking up certain substances and rejecting others. The phenomenon in its whole extent is probably a mechanical one; the great tendency of the salts of magnesia to form crystals with the salts of the alkalies, may probably in some measure impede their free solution and resorption.

Berzelius found 12·9% of phosphate of magnesia, and 25·8% of phosphate of lime in the ash of the excrements, after the use of coarse bread and a little animal food. Fleitmann<sup>2</sup> found that, after the use, for some days, of a diet consisting of more animal than vegetable food, the excrements yielded an ash containing 10·67% of magnesia.

The common intestinal concretions of horses consist almost entirely of phosphate of magnesia and ammonia, with fragments of straw, &c.; in a concretion of this sort, Simon<sup>3</sup> found 81% of phosphate of magnesia, but no salt of lime.

Physicians have paid much attention to the crystals of phosphate of magnesia and ammonia, which are very strikingly seen in typhous stools. Although these crystals are often enough to be found in the *feces* in other diseases, it must be granted that their occurrence is by far the most frequently to be noticed in abdominal typhus; indeed, it is well known that the ulcerated patches of the intestine are usually thickly studded with minute crystals of this nature.

Phosphate of magnesia is always found in the urine of man and of carnivorous animals, and its presence is rendered very perceptible when the urine becomes alkaline, by the readiness with which it crystallizes in combination with ammonia. As we shall return to this subject in the second volume, it is sufficient to observe in the present place, that these crystals are always formed in normal urine when alkaline fermentation commences. In serious lesions of the bladder or the spinal cord, we often find whole sediments consisting of these crystals. These deposits are, for the most part, either devoid of color, or of a dirty white tint. In a specimen of diabetic urine, I once found a glistening white sediment, consisting entirely of these crystals, and not containing a trace of lime. Urinary calculi, consisting of pure phosphate of magnesia, are very rare, although more common than the *fusible* calculi, which are

<sup>1</sup> Lehrb. d. Ch. Bd. 9, S. 345.

<sup>2</sup> Pogg. Ann. Bd. 76, S. 383.

<sup>3</sup> Buchner's Repertorium. Bd. 16, S. 215.

composed of a mixture of phosphate of lime with phosphate of ammonia and magnesia.\*

### FLUORIDE OF CALCIUM.

It is only in very minute quantities that this body occurs in the animal organism; it is, however, so integral a part of the enamel of the teeth, that we are inclined to ascribe to its presence (at least in part) the polish and the extraordinary hardness of that substance. The presence of small quantities of fluoride of calcium has been determined with certainty in the bones of almost all animals. More fluoride of calcium has been found in the skeletons of fossil animals than in those of our own time; and it is worthy of notice, that human bones found at Pompeii, contain, according to Liebig,<sup>1</sup> more fluoride of calcium than recent human bones.

Berzelius<sup>2</sup> found 2.1% of fluoride of calcium in the dentine and 3.2% in the enamel of a man's tooth, while the dentine and the enamel of that of an ox contained respectively 5.69% and 4% of this constituent. Marchand<sup>3</sup> found 1% in the femur of a man aged 30 years, and Heintz,<sup>4</sup> 2.05%.

Both Middleton<sup>5</sup> and von Bibra<sup>6</sup> have very carefully analyzed the bones of various classes of animals, and have recognized the presence of fluoride of calcium not only in the bones of the mammalia, but also in those of birds, fishes, and reptiles, and even in the shells of the mollusca. Middleton's assertion that the bones of a 6½ months' foetus contains as much fluoride of calcium as those of an adult, must be regarded as doubtful, till confirmed by further experiments.

Fluoride of calcium was first discovered in fossil ivory by Morichini;<sup>7</sup> it has since been found in all fossil bones by Proust,<sup>8</sup> Fourcroy and Vauquelin,<sup>9</sup> Chevreul,<sup>10</sup> Brandes,<sup>11</sup> Bergemann,<sup>12</sup> Marchand, von Bibra, Middleton, and others. Lassaigne<sup>13</sup> found as much as 15% in the teeth of an *Anoplotherium*, and I<sup>14</sup> found 16% in the outer portion of one of the ribs of the *Hydrarchos*.

[The presence of fluorine in blood and milk has been clearly demonstrated by Dr. George Wilson.<sup>15</sup>—G. E. D.]

In regard to the origin of the fluoride of calcium we cannot doubt that the small quantities found in the animal body may be easily conveyed into the system with the food; we need only remember that many mineral waters contain traces of fluorides, and that plants take up a little fluoride of calcium from micaceous soils.

Fluoride of calcium was detected by Berzelius in the Carlsbad water, and has been found in other mineral waters; moreover, artificially prepared fluoride of calcium is by no means perfectly insoluble in distilled

<sup>1</sup> Organ. Ch. auf Agricultur u. Physiol. angewendet, 1840, S. 140.

<sup>2</sup> Alt. Gehlen's Journ. Bd. 3, S. 1.

<sup>3</sup> Journ. f. pr. Ch. Bd. 27, S. 83.

<sup>4</sup> Ber. d. Ak. d. Wiss. z. Berlin. Febr. 1849, S. 51.

<sup>5</sup> Philos. Mag. T. 25, p. 14.

<sup>6</sup> Op. cit.

<sup>7</sup> A. Gehl. J. Bd. 3, S. 625; N. Gehl. J. Bd. 2, S. 177.

<sup>8</sup> N. Gehl. J. Bd. 2, S. 187.

<sup>9</sup> Ann. d. Chim. T. 57, p. 37.

<sup>10</sup> Ibid. p. 45.

<sup>11</sup> Schweigg. Journ. Bd. 32, S. 505.

<sup>12</sup> Ibid. Bd. 52, S. 145.

<sup>13</sup> Journ. de Pharm. T. 7, p. 1.

<sup>14</sup> Carus, über den *Hydrarchos*. Dresd. 1846.

<sup>15</sup> Edin. New Phil. Journ. Oct. 1850.

water. [According to Wilson 16 fluid ounces, or 7000 grains of water at 60° F., dissolve 0·26 of a grain of fluor spar.—G.<sup>1</sup> E. D.]

Whether the large quantities of fluoride of calcium which have been found in fossil bones are solely due to infiltration from without, must remain for the present undecided.

## SILICA.

As the skeleton of the vertebrate animals chiefly owes its hardness to the phosphate of lime which it contains, and the shell of the invertebrate animals to the carbonate of lime, so the shields of the lowest classes of animals are rendered hard and firm by containing a large quantity of silica. This substance is so thickly deposited in these organs that neither decomposition nor incineration can destroy their form; hence it is that deposits of fossil infusoria are so often discovered.

Silica for the most part occurs only as an incidental constituent of the juices and tissues of the higher classes of animals; Gorup-Besanez<sup>1</sup> has, however, shown by numerous experiments that this body forms an integral constituent of *feathers* and of *hair*.

Small quantities of silica have also been found in the blood, in the white of egg, in the bile, in urine, and in the solid excrements, and occasionally in certain morbid concretions.

The *Bacillariæ* are the most remarkable of all the infusoria in relation to the quantity of silica which they contain; their shields equally resist the action of fire and of acids. We are indebted to Ehrenberg<sup>2</sup> for our first accurate knowledge on this subject, and for the discovery of fossil infusoria in flint, mountain meal, &c.

Henneberg,<sup>3</sup> as well as Gorup-Besanez, has determined the quantity of silica in feathers; the latter, however, has fully investigated the subject in all its bearings, and extends his inquiry to the determination of the influences exercised by species, age, food, and other circumstances, on the deposition of silica in the feathers.

Gorup generally found from 0·11 to 2·47% of silica in the feathers of different birds, and from 6·9 to 65·0% of silica in the ash. The last-named quantity, which was the largest he ever found, occurred in the feathers of *Perdix cinerea*, but the feathers of *Strix flammea*, *Gallus domesticus*, and *Corvus frugilegus*, yielded ashes very rich in silica. The feathers of granivorous birds contained from 1·69 to 3·71% of silica (and their ash yielded from 25·5 to 50%); the feathers of birds living on fish and aquatic plants contained on an average 0·23%, and their ash 10·5% of silica; those of birds living on flesh and insects yielded, as a mean, 0·64%, and their ash 27%; and those of birds living on insects and berries 0·75%, and their ash 27%. Gorup usually found about twice as much silica in the feathers of old animals as in those of the young of the same species.

In newly grown or young feathers, only traces of silica were often to be found. In the pinions of the first order there was twice as much

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 66, S. 321-342.

<sup>2</sup> Die Infusionsthierchen u. s. w. S. 143-169.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 255-61.



silica, as in the tail-feathers of the second order; and in the tail and breast-feathers there was little more than in the pinions of the second order.

Berzelius found no silica in the *bones* or *teeth* of man; Fourcroy and Vauquelin<sup>1</sup> have, however, found it in the bones of children, and Marchand<sup>2</sup> in those of *Squalis cornubicus*; it has also frequently been found in fossil bones.

Silica has been found by Chevreul<sup>3</sup> in sheep's wool, and by Vauquelin,<sup>4</sup> and more recently by Laër,<sup>5</sup> in the human hair. Gorup has entered very fully into this part of the inquiry regarding the occurrence of silica. In brown human hair he found 0.22% of silica, the ash yielding 13.89%, while in the hair and wool of various animals he found sometimes rather more, and sometimes rather less of this substance. The quantity of silica in the hair appears to be altogether independent of the nature of the food.

As silica occurs so constantly in the animal organism, it might naturally be expected that we should find it in the *blood*, and especially in that of birds. Millon<sup>6</sup> found it in human blood; Weber<sup>7</sup> found that it amounted to 0.19%, in the ash of ox-blood, and in hens' blood Henneberg<sup>8</sup> found 0.96%.

Poleck<sup>9</sup> found 7.05%, in the ash of the *white of egg*; silica has also been found in the *bile, urine, and solid excrements*. Weidenbusch<sup>10</sup> found 0.36% in the ash of ox-bile; Pleisch<sup>11</sup> and Bley<sup>12</sup> detected it in gall-stones, and Mitscherlich<sup>13</sup> found a trace of it in the saliva. Berzelius<sup>14</sup> was the first who discovered traces of it in human urine; Fleitmann<sup>15</sup> has since found it in the ash of the urine, and Fourcroy and Vauquelin,<sup>16</sup> as well as De Koninck and Wurzer,<sup>17</sup> in urinary calculi. It need cause no wonder that silica is often found in the contents of the intestines, as it is widely distributed throughout the vegetable kingdom.

That the quantity of silica occurring in the animal organism essentially depends on the greater or lesser quantity of silica in the food, and consequently, that the origin of this body must be principally referred to vegetable food and siliceous water (and further, perhaps, in the case of birds, to the sand which they swallow), is rendered sufficiently evident from the experiments of Gorup-Besanez, if, indeed, any demonstration of the fact were required.

Plants contain far more silica than was formerly supposed; in the *Equisetaceæ*, for instance, the ash often contains 97%. The best method of exhibiting its presence in the seeds of the grasses, is by moistening them with a little nitric acid before incinerating them; in this manner, and with the aid of the microscope, we may, according to Schultz, recog-

<sup>1</sup> Ann. de Chim. T. 72, p. 282.

<sup>2</sup> Compt. rend. T. 10, p. 632.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 44, S. 172.

<sup>4</sup> Journ. de Pharm. 3 Sér. T. 13, pp. 86-88.

<sup>5</sup> Pogg. Ann. Bd. 76, S. 387.

<sup>6</sup> Pogg. Ann. Bd. 76, S. 360.

<sup>7</sup> Kastn. Arch. Bd. 8, S. 300.

<sup>8</sup> Pogg. Ann. Bd. 26, S. 320.

<sup>9</sup> Pogg. Ann. Bd. 76, S. 358.

<sup>10</sup> Schweig. Journ. Bd. 36, S. 321.

<sup>11</sup> Lehrb. d. phys. Ch. S. 97.

<sup>12</sup> Ann. de Chim. T. 58, p. 41.

<sup>13</sup> Op. cit.

<sup>14</sup> Ibid. S. 369.

<sup>15</sup> Journ. f. pr. Ch. Bd. 1, S. 115.

<sup>16</sup> Lehrb. d. Chem. Bd. 9, S. 433.

<sup>17</sup> Syst. des. Connoiss. Chim. T. 10.

nize the presence of this substance, not only in the husks, but also in the ovaries of many of the monocotyledons. Hence, it is obvious, that we must receive silica into the system with the bread; we can thus readily understand how it was that, after the use of rye-bread, Berzelius<sup>1</sup> found 1.016% in the solid excrements, and why it is that the dung of the *herbivora* (whose food consists of those parts of plants which are richest in silica), contains so large a quantity of this substance. In the dung of the cow, Zierl<sup>2</sup> found 4.4%, in that of the sheep, 6.0%, and in that of the horse, 4.6%. Hence, large quantities of silica are often found in the intestinal concretions of herbivorous animals.

## SECOND CLASS OF MINERAL BODIES.

### HYDROCHLORIC ACID.

As we are convinced by the reasons given in p. 93, that lactic acid is the essential free acid of the gastric juice, we need devote no special consideration to this acid. It is sufficient to remind our readers, that, according to our experiments,<sup>3</sup> lactic acid can be replaced by no other acid, except hydrochloric acid, in the process of digestion.

### HYDROFLUORIC ACID.

Brugnatelli<sup>4</sup> believed that he had discovered the existence of this acid in the gastric juice of birds, when he found that pieces of agate and rock-crystal, which he introduced by means of tubes into the stomachs of common fowls and turkeys, were distinctly corroded, and had lost from 12 to 14 grains in weight, on their removal after ten days; and Treviranus<sup>5</sup> also believed that, when the contents of the intestinal canal of fowls were digested in porcelain vessels, the glazing was attacked.

In reference to the small quantities of this acid which might possibly occur in the gastric and intestinal juices of these animals, it is certainly difficult to demonstrate its absence in an unquestionable manner; but as theoretical reasons as well as direct experiments are opposed to Brugnatelli's view, we may, at all events, with great probability, assume the non-occurrence of this acid. Tiedemann and Gmelin,<sup>6</sup> digested the gastric juice of a duck for 24 hours in a platinum crucible, which was covered with a piece of glass, having a coating of wax through which a few lines were drawn; they could, however, detect no corrosion on the glass. I placed the chyle of a duck which had just been killed, in a platinum crucible, treated the mass with a little sulphuric acid, and covered the crucible with a watch-glass coated with wax, except at the centre (the inferior convex part), where its surface was bare and exposed; at the termination of the experiment, I could not find the slightest corrosion on the watch-glass. Further, I saturated with potash the fluid

<sup>1</sup> Lehrb. d. Chem. Bd. 9, S. 346.

<sup>2</sup> Kastn. Arch. Bd. 2, S. 476.

<sup>3</sup> Ber. d. k. Sächs. Ges. d. Wiss. z. Leipzig. 1849.

<sup>4</sup> Crell's Ann. 1787. Bd. 1, S. 280.

<sup>5</sup> Biologie Bd. 4, S. 862.

<sup>6</sup> Verdauung. Bd. 2, S. 139.

obtained by washing the contents of the crop and stomach of two turkeys with water, evaporated it to dryness and burned the residue; the ash was then carefully treated with sulphuric acid in a platinum crucible, in the manner already described, but here also no trace of hydrofluoric acid was obtained.

If these experiments are not sufficiently stringent to overthrow the observations of Brugnatelli, they, at all events, serve to explain how it was that Brugnatelli and Treviranus were led to adopt this view. For it is very possible that, as we always find small pebbles and sand in the stomachs of these animals, a purely mechanical attrition of the finest granules of sand may have apparently corroded the pieces of agate and rock-crystal during their long sojourn in the stomach, and thus have occasioned their loss of weight. Moreover, I have never been able to detect any decided corrosion of the pebbles, which we find in the stomachs of ducks and fowls. It would be strange if nature had here first ordained the secretion of hydrofluoric acid, in order that it should immediately again disappear through the action of the siliceous pebbles which are swallowed by birds. Should not the hydrofluoric acid, if it were present, expel other acids from the salts contained in the gastric juice?

#### CHLORIDE OF SODIUM.

In almost every portion of the earth's surface we find this body in all parts of the animal organism; and it is not a mere incidental constituent conveyed into the system with the food and drink, but it is applied to definite, although highly various ends.

The importance of chloride of sodium in the metamorphosis of the animal tissues is illustrated by the fact that it always forms the greatest part of the soluble constituents of the ash of all animal substances. It is very constantly associated with certain animal matters, and essentially influences their chemical and physical properties; thus albumen in part owes its solubility to the chloride of sodium contained in it, and the differences which it presents in coagulating are, in part, dependent on the quantity of this salt that is present. Chloride of sodium dissolves pure casein, and has a singular power of impeding the coagulation of the fibrin of the blood. If it is impossible to prove that chloride of sodium forms definite chemical compounds with these bodies, the following considerations, at all events, render such a view probable;—namely, the influence this salt exercises on the above-named protein-compounds, the analogy of the compound of chloride of sodium and glucose, and finally, the impossibility, by mere washing, of perfectly separating some of the protein-compounds from the chloride of sodium.

We would especially refer the reader to the relation of albumen towards salts, described in p. 296.

In accordance with these facts we find that the chloride of sodium, like other important constituents of the animal body, is not merely constantly present, but also that it is combined in tolerably definite proportions in the different constituent parts. For it is an established law, that the different animal fluids always strive to attain a similar chemical

constitution. This law, to which we must subsequently recur more in detail, includes the protein-compounds, which, if they are taken in excess, certainly are decomposed in the ordinary manner, but are eliminated as rapidly as possible by the kidneys, under the form of urea and uric acid.

The chloride of sodium in normal human blood stands in a tolerably constant ratio to its other soluble constituents, the limiting ratios being 3 : 1 and 2·4 : 1. Berzelius<sup>1</sup> found 6 parts in 1000 of the serum of human blood, and Marcet<sup>2</sup> 6·6 in 1000 parts of blood, which corresponds to about 5·5 in 1000 of serum; Nasse<sup>3</sup> obtained from 4 to 5 parts of chloride of sodium from 1000 of blood, Denis<sup>4</sup> from 3·537 to 3·668 parts, and Becquerel and Rodier<sup>5</sup> from 2·3 to 4·2 parts; the mean of 11 analyses of men's blood yielding 3·1, and of 8 analyses of women's blood 3·5 parts. In 1000 parts of my own blood in a normal state I found 4·138 parts of chloride of sodium, and after the use of very salt food, which caused intense thirst, it amounted to 4·148: an hour after taking two ounces of salt, and having in the interval drank about two quarts of water, the quantity was 4·181. Hence it seems to follow that the animal organism not only removes foreign substances with extraordinary rapidity, but that even useful substances, if they are in excess, are as rapidly as possible eliminated.

The amount of salt in the blood undergoes great fluctuations in different diseases; thus Nasse<sup>6</sup> and Scherer<sup>7</sup> found that there was a diminution of the chloride of sodium in inflammatory blood; O'Shaugnessy, Rayer, and Mulder observed this strikingly in cholera; Nasse also observed it in the blood of a diabetic patient, Lecanu in cases of jaundice, and Jennings and Simon in chlorotic patients: an augmentation of the salt in the blood has been noticed by Fremy in sea-scurvy and by Nasse in the rot in sheep. My experiments have left it very doubtful whether the salts of the blood are diminished in tuberculosis, since it is not often that we can obtain the blood of tuberculous patients, except when some inflammatory attack gives occasion for the abstraction of blood. We shall return to this subject more fully in the second volume, when considering "The Blood."

Even if the well-known action of chloride of sodium on the color of the blood be entirely dependent on mechanical relations, the occurrence of almost constant quantities of this salt in the blood during health, and its considerable variations in different diseases, and, further, its chemical action on histogenetic substances, indicate that in all probability it takes some definite chemical part in the metamorphosis of the blood. Hofmann<sup>8</sup> believes that it increases the capacity of the constituents of the blood for oxidation, which, however, requires proof.

Berzelius was formerly of opinion that the quantity of albumen contained in the serum of the blood might be the cause why the blood-pigment which is so readily soluble in pure water did not dissolve in the serum, but Joh. Müller has shown that the capsules of the blood-cor-

<sup>1</sup> *Lehrb. d. Chem.* Bd. 9, S. 98.

<sup>2</sup> *Handwörterbuch d. Physiol.* Bd. 1, S. 167.

<sup>3</sup> *Gaz. méd.* 1844, No. 48.

<sup>4</sup> *Haeser's Arch.* Bd. 10.

<sup>5</sup> *Medico-Chir. Trans.* Vol. 2, p. 370.

<sup>6</sup> *Journ. de Chim. méd.* T. 4, p. 111.

<sup>7</sup> *Das Blut.* 1836, S. 287.

<sup>8</sup> *Das Protein u. s. w.* S. 19.

puscles dissolve, if they are brought in contact with an aqueous and not too dilute solution of albumen; if, however, we treat the albumen with a little water containing only 1% of chloride of sodium, the corpuscles remain unchanged, whereas they are destroyed by a pure solution of salt containing no albumen.

We shall treat, at some length, of the mode of action of chloride of sodium and various other bodies on the red color of the blood, in the second volume. It is here sufficient to remark that Scherer's experiments have clearly demonstrated that the bright or dark color of the blood principally depends on the form of the blood-corpuscles, which again is chiefly dependent on the endosmotic relations existing between their contents and the surrounding fluid. For instance, if we add much salt to blood, the corpuscles become contracted and biconcave: it is to this biconcave form that Scherer attributes the brighter color of the blood.

In those fluids which are secreted from the blood and which contain a larger quantity of chloride of sodium than the blood itself, as, for instance, the saliva, gastric juice, inflammatory exudations, pus, and mucus, this salt doubtless discharges some important functions. We claim no high importance for it in the saliva; but if that fluid exercises a function, the chloride of sodium certainly takes part therein, since its quantity exceeds that of all the other constituents of the saliva. In the gastric juice we find, in addition to a little organic matter, scarcely anything but metallic chlorides, and chiefly chloride of sodium. From the abundance in which it exists both in the saliva and the gastric juice we might be led to infer that it essentially promotes the solution of the food, and its future changes, or at all events, that it contributes to impede abnormal decompositions and metamorphoses of the food.

Several observations which I have made, tend to show that the excess of salt conveyed into the blood is not merely carried off by the kidneys with the greatest possible rapidity, but also by other secreting organs, as the salivary glands, the gastric glands, &c. While the gastric mucous membrane of a dog with a fistulous opening into the stomach, secreted a juice when the stomach was empty and artificially stimulated, which, according to Blondlot, contained 0.126% of chloride of sodium, I obtained a gastric juice in a similar manner from a dog into whose jugular vein I had half an hour previously injected two ounces of a saturated solution of salt, which contained 0.385%. These facts are rendered more perceptible by using either of the analogous salts, the iodide of sodium, or of potassium; iodide of potassium, when injected into the veins, appears with extreme rapidity in the stomach, although I am not quite certain whether this is not in a great measure dependent on its very rapid presence in the saliva, and on its finding its way into the stomach through that fluid; for I have convinced myself that the iodide of potassium passes from the blood in larger quantity, and with more rapidity, into the saliva than into the urine. If we take a few grains of iodide of potassium in the form of pills, and at once convince ourselves that no iodine is retained in the buccal fluids, we can in the course of from 5 to 10 minutes recognize iodine with certainty in the saliva, although it cannot be then detected in the urine even if we examine that fluid directly after its

secretion by the kidneys, as it drops from the ureters. Bernard<sup>1</sup> has made similar observations with prussiate of potash, lactic acid, and other substances; after injection into the jugular veins of a dog, they very rapidly appeared in the gastric juice.

Enderlin<sup>2</sup> found 61.93% of the chlorides of sodium and potassium in 100 parts of the mineral constituents of saliva.

Prout<sup>3</sup> found from 0.12% to 0.13% of the chloride of sodium with a little chloride of potassium in human gastric juice; Braconnot,<sup>4</sup> Tiedemann and Gmelin,<sup>5</sup> and Berzelius agree in stating that the gastric juice is rich in this salt. I found 0.311% of chloride of sodium in the fluid from the crop of a duck which for eight days had been only fed with barley moistened with distilled water.

That the chloride of sodium, and the metallic chlorides generally, which are contained in the gastric juice, contribute at all to the solution of the histogenetic substances is not probable; for, notwithstanding some of my earlier experiments which seemed to support that view, more recent and more numerous experiments<sup>6</sup> have convinced me that any addition of salt, either to natural or well-prepared artificial gastric juice, infallibly retards the changes which the articles of nitrogenous food undergo. We may presume that a definite quantity of the metallic chlorides exists in some form of chemical combination in the gastric juice; this quantity being exactly sufficient to hinder any abnormal decomposition in that fluid, without checking its digestive power.

In the exudations we certainly find less chloride of sodium than in the blood itself, but in relation to the fixed constituents of these liquids, this salt is always considerably increased. The investigations of Brucke<sup>7</sup> and Henle<sup>8</sup> have proved, almost beyond a doubt, that this abundant transudation of soluble salts through the walls of the vessels is dependent on a purely mechanical relation. It is, however, not improbable that the chloride of sodium co-operates in the metamorphosis of the exudation; we find, at least, that pus and other exudations in which cells become developed, are very rich in this salt; and this is especially the case with mucus, as has been shown by Nasse.<sup>9</sup> The fluid of cancerous growths always contains a large quantity of this salt. Whether the chloride of sodium takes part in the abnormal conversion of the exudation into cells, is a question that must be at present left undecided. We are almost led to the belief that every deposition of cells is accompanied by an increase in the quantity of chloride of sodium, or that this salt arrests their development at a low stage. We find, at least, that the cartilages, which, in their perfectly developed state abound in cells, contain far more chloride of sodium than occurs in other parts of the animal body. The cartilaginous bones of the foetus, before much phosphate of lime has been deposited, contain far more chloride of sodium than adult bones: and abnormal depositions of bony matter contain more of this salt than even the permanent cartilages.

<sup>1</sup> Thèse soutenue à la faculté de Paris, 1844.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 50, S. 56.

<sup>4</sup> Ann. de Chim. et de Phys. T. 59, p. 113.

<sup>5</sup> Verdauung. Bd. 1, S. 91.

<sup>7</sup> Casper's Wochensch. 1840, No. 21.

<sup>9</sup> Journ. f. prakt. Ch. Bd. 29, S. 59.

<sup>3</sup> Phil. Trans. for 1824, p. 45.

<sup>6</sup> Ber. d. k. sächs. Ges. d. Wiss. 1849.

<sup>8</sup> Zeitschr. f. rat. Med. Bd. 1, S. 122.

Fromherz and Gugert<sup>1</sup> found 8·231% of chloride of sodium in the ash of the costal cartilages of a man aged 20 years; I found 11·236% of this salt in the ash of the laryngeal cartilages of an adult female. From various bones I could only extract from 0·7 to 1·5%. The femur of a six-months' foetus which I examined contained 10·138% of chloride of sodium, and according to Valentin<sup>2</sup> the incrusting exudation, deposited around a carious tibia, contained 13·7%.

Nasse, taking the mean of two analyses, found that the chloride of sodium in the mucus of the air-passages amounted to 0·582%, while two comparative analyses showed that it amounted to 0·46% in the serum of the blood, and to 1·26% in that of pus. Hence in this respect pus approximates closely to mucus, while the serous portions of blood and pus are differently constituted.

In order to give a general view regarding the occurrence of chloride of sodium in the animal fluids, I append the following table, which is based; in a great measure, on my own analyses; *a* signifies the amount of salt in 100 parts of the fluid, *b* in 100 parts of solid residue, and *c* in 100 parts of ash.

	<i>a.</i>	<i>b.</i>	<i>c.</i>
Human blood, . . . . .	0·421%	1·931%	57·641%
Blood of the horse, . . . . .	0·510%	2·750%	67·105%
Chyle, . . . . .	0·531%	8·313%	67·884%
Lymph (Nasse), . . . . .	0·412%	8·246%	72·902%
Serum of the blood (Nasse), . . . . .	0·405%	5·200%	59·090%
Blood of the cat (Nasse), . . . . .	0·537%	2·826%	67·128%
Chyle (Nasse), . . . . .	0·710%	7·529%	62·286%
Human milk, . . . . .	0·087%	0·726%	33·089%
Saliva, . . . . .	0·153%	12·988%	62·195%
Gastric juice of the dog, . . . . .	0·126%	12·753%	42·089%
Human bile, . . . . .	0·364%	3·353%	30·464%
Urine, . . . . .	0·332%	5·187%	22·972%
Mucus (Nasse), . . . . .	0·583%	13·100%	70·000%
Serum of the blood (Nasse), . . . . .	0·460%	4·919%	58·974%
Serum of pus (Nasse), . . . . .	1·260%	11·454%	72·330%
Inflammatory exudation in the pleura (Scherer), . . . . .	0·750%	10·416%	73·529%
Scirrhus of the breast, . . . . .	0·314%	6·043%	65·391%

After this general view of the occurrence and uses of salt in the animal economy, it is hardly requisite to allude to the sources from which the animal body receives its due supply. Chloride of sodium is so generally distributed throughout nature, that this necessary quantity is conveyed into the organism with the ordinary food and with the water.

The habits of civilized life have elevated salt to the rank of a positive necessary, but we must by no means conclude from this circumstance that the salt contained in ordinary food is not sufficient for the support of the animal functions. A simple comparison of the quantity of salt contained in the animal body, with that which we are daily taking with the food, at once shows that we use more salt than is requisite; and if, on the one hand, as several travellers narrate, certain negro tribes in the interior of Africa exchange gold-dust for an equal weight of salt, and in want of it have recourse to the most disgusting substitutes; we know, on the other hand, that whole races in the South Sea Islands, and in

<sup>1</sup> Schweigg. Journ. Bd. 50, S. 187.

<sup>2</sup> Reporter. 1838, S. 301.

South America, flourish without even the knowledge of this substance. Further, as Liebig has shown, tempests carry salt from the ocean far into the interior, and thus supply the spring water with it. A glance at the results of the analyses of the ashes of plants, is sufficient to show that the ordinary articles of vegetable food are perfectly sufficient to supply the necessary quantity of salt to the animal body.

### CARBONATE OF SODA.

This salt not unfrequently occurs in the ash of burned animal matters, but in most cases it is merely the product of the combustion of combinations of soda with organic acids or protein-compounds. Investigations deserving of the greatest confidence prove however that carbonate of soda, together with other soda-compounds, exists in the blood and in the lymph. It is also contained, together with large quantities of the carbonate of potash and lime, in the urine of herbivorous animals.

The earlier observers assumed the presence of carbonate of soda in the blood as a recognized fact; and indeed it was believed to take an active part in the excretion of carbonic acid; but certain later investigations seemed to leave it very doubtful whether alkaline carbonates exist in the blood. Alkaline carbonates were always found in the ash of blood (as for instance, by Berzelius, Marcet, Mitscherlich, Tiedemann and Gmelin, and more recently by Nasse, Marchand, and others), till Enderlin<sup>1</sup> announced that blood incinerated according to this method, left an ash which did not yield a trace of carbonic acid. He examined the ash of the blood of men, oxen, sheep, and hares, and found that in addition to the ordinary chlorides and sulphates, the soluble salts consisted solely of tribasic phosphate of soda. Hence he concludes that as no carbonates can be found in the ash, it is altogether impossible that any carbonated alkali can occur in the blood. But it does not follow that the earlier observers were in error, when they found carbonate of soda in the blood (Nasse,<sup>2</sup> for instance, found from 0.06 to 0.08%, and Marchand,<sup>3</sup> 0.125%), for we can at pleasure prepare a blood-ash either with or without carbonates, according to the degree of heat and the method of incineration we employ. If we heat common phosphate of soda ( $2\text{NaO} \cdot \text{HO} \cdot \text{PO}_5$ ) with carbonate of soda, the latter loses its carbonic acid, and as a necessary consequence there is formed the tribasic phosphate of soda; when dissolved in water, this tribasic phosphate of soda very rapidly absorbs carbonic acid from the atmosphere, and becomes converted into carbonate and *c* (common) phosphate of soda. Hence tribasic phosphate of soda cannot exist in the circulating blood, since this fluid contains sufficient carbonic acid to insure its decomposition.

Assuming that carbonate of soda exists in the blood-ash, this by no means proves that it is present in fresh blood, for this fluid contains fatty and other organic acids in combination with alkalis, which on incineration are converted into carbonates. But if we consider that fresh blood

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 50, S. 53. <sup>2</sup> Handwörterb. der Physiolog. Bd. 1, S. 167.

<sup>3</sup> Lehrb. d. physiol. Chem. S. 226.



always has an alkaline reaction, and that, in consequence of its always containing carbonic acid, caustic soda can no more occur in it than the above-mentioned tribasic phosphate of soda, this reaction can hardly be attributed to any other body than to carbonate of soda; for the combinations of the fatty acids with alkalis are contained in the blood in far too small quantities to account for the alkaline reaction of that fluid, and the amount of carbonate present in the ash. Liebig<sup>1</sup> was the first to remark that the carbonate of soda must be contained in the blood as a bicarbonate. No free acid can be present with common carbonate of soda. The following experiment favors the view of the presence of the bicarbonate: if we precipitate the serum of the blood with alcohol and thoroughly wash the precipitate with dilute spirit, the albumen on incineration leaves no alkaline ash; if soda were chemically combined with albumen, the soda must be precipitated with the albumen, while neutral carbonate of soda and especially the bicarbonate dissolve readily in spirit. On passing hydrogen through the fluid from which the albumen has been removed by filtration, carbonic acid is expelled; for as Magnus and Rose formerly proved, and as Marchand<sup>2</sup> has recently again demonstrated, hydrogen completely expels the one atom of carbonic acid from the bicarbonate of soda, especially if the temperature be raised to 38°. Liebig also adduces the relation of corrosive sublimate to the fluid freed from the albumen by spirit of wine, in evidence of the presence of bicarbonate of soda; for, on the addition of corrosive sublimate to this fluid, there is no precipitate, but after some time there are deposited brown crystals of oxychloride of mercury, precisely as would have occurred if this reagent had been added to a solution of bicarbonate of soda. By means of a current of pure hydrogen gas, and by the repeated application of the air-pump, I so thoroughly removed the carbonic acid from freshly whipped ox-blood, that a fresh stream of hydrogen passed through the blood no longer produced the slightest turbidity in baryta-water; by means of a special contrivance, so as to exclude the access of the air, a little acetic acid was forced into the blood by means of the hydrogen gas, and the latter was again passed in considerable quantity through the blood; immediately after the addition of the acetic acid to the blood the baryta-water was rendered turbid by the current of hydrogen. We thus obtain a proof that a certain quantity of the carbonic acid in the blood exists in combination with a base, in addition to that which can be expelled by gases and extracted by the air-pump. Hence there can no longer be any doubt regarding the presence of carbonate of soda in the blood. I have found, taking the mean of ten carefully conducted quantitative analyses,<sup>3</sup> that ox-blood contains 0.1628% of ordinary carbonate of soda, after the expulsion of the free carbonic acid in the manner which has already been described.

Nasse<sup>4</sup> found 0.056% of carbonate of soda in the lymph of a horse, while Marcet<sup>5</sup> found 0.165% in the serum of the blood. Those who re-

<sup>1</sup> Handwörterb. der Chem. Bd. 1, S. 901.

<sup>2</sup> Journ. f. pr. Chem. Bd. 35, S. 390.

<sup>3</sup> Ber. d. k. sächs. Ges. d. Wiss. 1847, S. 96-100.

<sup>4</sup> Simon's Beiträge z. phys. u. pathol. Chem. Bd. 1, S. 449.

<sup>5</sup> Medico-Chir. Trans. vol. 2, p. 370.

gard the kidneys as mere percolators cannot deny the presence of alkaline carbonates in the blood, since the urine (at least of herbivorous animals) contains a considerable amount of carbonates. The parotid saliva of the horse becomes turbid, in the same manner as lime-water, on exposure to the air, with, however, this difference, that it almost immediately deposits the most beautiful microscopic crystals of carbonate of lime.

Liebig was formerly of opinion that the carbonate of soda in the blood acted an extremely important part in the process of respiration, in short, that it was the means by which the carbonic acid is conveyed from the capillaries into the lungs. The oxygen mixed with the blood in the lungs there displaces the carbonic acid as completely as it would be expelled by a current of oxygen or hydrogen from its state of combination in bicarbonate of soda. As far as our present knowledge extends, no facts are at variance with this view; indeed if the presence of carbonate of soda in the blood be once granted, no one can wonder that it is converted to the bicarbonate, and on the other hand, that it must be decomposed on coming in contact with other gases than carbonic acid. But the question naturally suggests itself—Is the quantity of carbonate of soda sufficient to serve as a means of transport for the whole of the carbonic acid of the blood? The following calculation supplies the answer: 1000 grammes of blood contain 1.628 grammes of carbonate of soda, which, to become converted into bicarbonate must take up 0.637 of a gramme of carbonic acid; hence 0.637 of a gramme of carbonic acid can be extracted from the blood by an air-pump, or expelled by other gases; this would amount to 322 cc. according to volume; if we assume that the specific gravity of the blood is 1.055, then 1000 cc. of blood would contain 343 cc. of carbonic acid, capable of being removed by other gases or by the air-pump. Magnus has, however, succeeded in removing about 300 cc. of carbonic acid from 1000 cc. of blood by means of hydrogen and a vacuum; a method by which a part of the carbonic acid must always remain in the blood. The coincidence between the empirical result and the calculation is quite as great as could be expected.

It cannot be doubted that the carbonate of soda in the blood serves as a solvent for the fibrin as well as the albumen; Bird has, however, shown that the bicarbonate is one of the best solvents for albumen. It is well known that large quantities of the alkaline carbonates have the property of impeding or altogether preventing the coagulation of the fibrin.

Finally, that the alkali of the blood also contributes to saturate the acids conveyed into the organism or formed within it, is the more probable, because nature seems to have provided that the alkaline carbonates shall be produced as rapidly as possible from the combinations of potash and soda with vegetable acids. (See p. 97.)

The origin of carbonate of soda in the animal body is so obvious, from the preceding observations, that it is unnecessary to enter further into the subject.

## ALKALINE PHOSPHATES.

Important as the alkaline phosphates doubtless are in the metamorphosis of animal tissue, we are unable at present to state much with certainty regarding them. Before Rose had introduced his new method of preparing and analyzing the ashes of organic bodies, it must have been concluded from the abundant occurrence of alkaline phosphates in the ashes of animal substances, that these salts played an important part in the animal economy. This conclusion seems especially to be supported by the peculiar relations of the saturating capacity of phosphoric acid, and by the metamerism of the phosphates. For it is almost self-evident that no salts of any other acid could be so usefully applied in the metamorphosis of tissue, as those of phosphoric acid, which can form neutral salts with one, two, and three atoms of base, acid salts with one and two atoms, and likewise several basic salts. Moreover it must be recollected that common phosphate of soda may contain one atom of basic water in place of one atom of fixed base, and thus by its alkalinity it may serve, like free alkalis or their carbonates, as a solvent for many animal substances;—that it has the property of yielding to the weakest acids, as, for instance, uric acid, one of the two atoms of fixed base, and of being converted into an acid phosphate;—and finally, that the ordinary basic phosphate of soda (with 3 atoms of fixed base) yields 1 atom of soda to free carbonic acid, and thus gives rise to two neutral salts, both of which, however, have an alkaline reaction, and a strong solvent power.

Taking all these circumstances into consideration, and moreover recollecting the importance of the earthy phosphates, and especially of the animal substances containing phosphorus, we might be disposed to believe the conclusion justified, which, it was supposed, might be drawn from the abundance with which alkaline phosphates occur in the ash. But, unfortunately, Rose's improved analyses of the mineral constituents occurring in animal bodies have deprived us of the basis on which this conclusion rests. The earlier ash-analyses of the different animal juices can no longer be regarded as affording evidence of the importance of these alkaline phosphates: later and more perfect analyses, in accordance with Rose's method, do not enable us to form a decided opinion regarding the occurrence of preformed alkaline phosphates in the different animal fluids, for it is not only the alkaline phosphates contained in the aqueous extract of the carbonaceous residue of animal bodies which are to be regarded as preformed in the animal body, but also those contained in the hydrochloric extract, which were retained in the residue with phosphate of lime, or of magnesia as insoluble double salts (Rose)<sup>1</sup>.

We cannot decide, in reference to these alkaline phosphates, whether previously to their combining with lime or magnesia, they existed preformed as basic alkaline phosphates, or rather, as Rose thinks more probable, as alkaline carbonates or combinations of alkalis with organic acids; further, it has never been quite accurately determined to what extent alkaline phosphates are produced from phosphate of magnesia

<sup>1</sup> Pogg. Ann. Bd. 77, S. 288–302.

when decomposed by alkaline carbonates. But putting out of view all these uncertainties, we should not be too hasty in drawing conclusions from the results of such analyses of the mineral constituents; for the principle asserted by Rose that the mineral bodies which cannot be extracted by hydrochloric acid from the carbonaceous residue of animal substances must be regarded as non-oxidized, and as combinations of phosphuretted radicals with metals, is at present only an hypothesis, although a very probable one. Such are the reasons which determine us for the present to suppress any consideration of the part which the alkaline phosphates may take in the general metamorphosis of matter, or in individual animal processes. If, however, further investigations demonstrate, with greater certainty, the more abundant occurrence of these phosphates in the individual animal juices and in certain processes, our knowledge of the properties of phosphate of soda, would readily lead us to understand in what manner the alkaline phosphates would act in the different processes.

In order to give some sort of general idea how, according to Rose's analyses, the preformed alkaline phosphates should stand in relation to the other mineral constituents, we have collected, in the following table, the results of the analyses of several animal substances, conducted under Rose's superintendence.

There are yielded by 100 parts of the ash of	Salts which can be extracted from the carbonaceous residue by water.	Alkaline phosphates contained in 100 parts of the soluble salts.
Ox-blood, . . . . .	60.90	$3\text{KO.PO}_5$ . . . . . 1.58
Horse-flesh, . . . . .	42.81	$\left\{ \begin{array}{l} 2\text{NaO.PO}_5 \\ 2\text{KO.PO}_5 \end{array} \right.$ . . . . . 11.10
Cow's milk, . . . . .	34.17	$\left\{ \begin{array}{l} 2\text{KO.PO}_5 \\ 3\text{KO.PO}_5 \end{array} \right.$ . . . . . 83.27
Yolk of egg, . . . . .	40.95	$\left\{ \begin{array}{l} \text{KO.PO}_5 \\ \text{NaO.PO}_5 \end{array} \right.$ . . . . . 21.60
White of egg, . . . . .	81.85	$\left\{ \begin{array}{l} \text{KO.PO}_5 \\ \text{NaO.PO}_5 \end{array} \right.$ . . . . . 24.57
Ox-bile, . . . . .	90.85	$\left\{ \begin{array}{l} \text{NaO.PO}_5 \\ 3\text{KO.PO}_5 \end{array} \right.$ . . . . . 25.16
Urine, . . . . .	90.87	$\left\{ \begin{array}{l} 3\text{KO.PO}_5 \\ 3\text{NaO.PO}_5 \end{array} \right.$ . . . . . 0.00
Solid excrements, . . . . .	18.55	$\left\{ \begin{array}{l} 3\text{KO.PO}_5 \\ 2\text{KO.PO}_5 \end{array} \right.$ . . . . . 6.78
		$\left\{ \begin{array}{l} 3\text{NaO.PO}_5 \\ 3\text{KO.PO}_5 \end{array} \right.$ . . . . . 14.51
		$\left\{ \begin{array}{l} 2\text{KO.PO}_5 \\ 3\text{KO.PO}_5 \end{array} \right.$ . . . . . 16.12
		$\left\{ \begin{array}{l} 3\text{KO.PO}_5 \\ 3\text{KO.PO}_5 \end{array} \right.$ . . . . . 4.55
		$\left\{ \begin{array}{l} 3\text{KO.PO}_5 \\ 3\text{KO.PO}_5 \end{array} \right.$ . . . . . 20.13

Even these few numerical results promise to throw much light on the theory of the metamorphosis of animal substances, on the nature of individual zoo-chemical processes, on the distribution of the potash and the soda in the different animal fluids, on the physiological importance of phosphorus, &c. Notwithstanding the confidence which we are justified in placing on the accuracy of these analyses, we avoid entering deeply into the conclusions that might be deduced from them, for independently of the circumstance that so few analyses afford us comparatively little means of establishing theories and deductions, we shall find sufficient occasion, when considering the animal substances named in the above table, to revert to the *data* afforded by these experiments, especially as our observations would extend to too great a length, if we were to attempt to bring into unison, or to estimate as they deserve, the often contradictory results of the earlier analyses.

Thus, for instance, in the consideration of the muscular tissue and of the fluid with which it is saturated, we shall enter into the beautiful views which Liebig, with his customary skill, has developed in his classical memoir on this subject. He has there particularly directed attention to the different proportions in which potash and soda exist in the blood and in the muscular fluid; this very important difference is less marked in Rose's analyses of the mineral constituents of both fluids, and taking into consideration the importance of the subject, it is exceedingly necessary, in order that we should have a clear insight into these relations, that we should form a decisive opinion regarding the value of the facts in our possession.

A glance at the numerous analyses of the ashes of plants, and especially of their seeds, is sufficient to indicate the *source* of the phosphates in the animal body; the copious discharge of phosphates by the urine need scarcely excite our wonder, as it includes both those which were contained preformed in the food, and those which are formed during the metamorphosis of animal tissue, by the oxidation of the phosphuretted organic matters or radicals.

### IRON.

This metal occurs in the animal body, not only in very different parts, but also in different conditions; in the blood, as we have already shown in our observations on hæmatin, it seems highly probable that it exists, for the most part, in a non-oxidized state; in the gastric juice it exists, according to Berzelius, as a protochloride, and in other fluids as a phosphate.

According to Rose's method, the ash of ox-blood contains 6.84% of peroxide of iron, that of horse-flesh 1.00%, that of milk 0.47%, that of the yolk of egg 1.85, that of the white of egg 2.09%, that of the bile 0.23%, and that of the fæces 2.09%. We have already noticed the presence of iron in black pigment in our remarks on melanin. Large quantities of iron are sometimes found in the ashes of gall-stones, especially of such as consist chiefly of pigment. There appears to be no relation between the color of the hair, and the quality of the iron which it contains (Laër.)<sup>1</sup>

We are unfortunately perfectly ignorant regarding the special uses of iron in the animal economy. In reference to the iron in the blood, we have already seen (p. 274) that it is in some way connected with the function of the corpuscles, but we know nothing further. But since the iron is of especial importance in the animal body, we cannot wonder at its occurrence in the milk and in the egg. If we find iron in the bile, its occurrence there is easily explained, if we adopt the view that this fluid is for the most part produced from the destruction of the blood-corpuscles.

The fluid and solid articles of food contain so much iron that a portion of it is always thrown off with the solid excrements. Nature has provided that the animal organism shall receive the necessary quantity of this essential metal with every kind of food.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 45, S. 227.

## THIRD CLASS OF MINERAL BODIES.

## ALKALINE SULPHATES.

Sulphates occur in most of the animal fluids, with the exception of the urine, in extremely small quantities; and, indeed, in several, as, for instance, the milk, the bile, and the gastric juice, they are altogether absent. They are also contained in comparatively minute quantities in the blood. Hence it may be concluded that these salts are of no essential use in the animal organism; a view which is confirmed by the fact that as soon as they are taken into the body, they are as rapidly as possible eliminated either with the solid or the fluid excrements. On the other hand, it is worthy of remark, that v. Bibra<sup>1</sup> found considerable quantities of soda in the bones of reptiles and fishes.

Berzelius<sup>2</sup> and Simon<sup>3</sup> found no sulphates in the milk, and Braconnot<sup>4</sup> and Berzelius<sup>5</sup> also failed to detect them both in the gastric juice, and in the bile of man and the ox.

If we treat the dry residuc of the serum of the blood, milk, saliva, bile, &c., with spirit, till it ceases to extract anything additional on boiling, and if we then extract the insoluble residuc with water, precipitate the aqueous solution with a little tannic acid, evaporate the filtered fluid, again extract with spirit, and dissolve the residuc in water, the aqueous solution only seldom exhibits any traces of sulphates. That sulphate of soda is frequently found even in considerable quantity in the ash of these animal fluids, and indeed that it must be found there, is sufficiently explained by the remarks we have already made regarding the changes which the mineral constituents of animal substances undergo on incineration. The bile presents one of the best examples of these changes, for its ash is very rich in sulphates, while we can hardly discover a trace of them in the fresh fluid.

The frequent use of the alkaline sulphates in medicine might almost lead to the presumption, that these salts when conveyed into the system with the food, are not devoid of use in relation to the physiological functions of the animal organism, and in particular to that of digestion. When on the one hand we take into consideration the changes which the alkaline sulphates undergo in the process of digestion, and, on the other the occurrence of highly sulphuretted organic substances in the animal organism, great probability seems to attach to this view. The experience of physicians, and direct physiologico-chemical experiments have clearly proved, that small quantities of alkaline sulphates are converted in the intestinal canal during digestion into sulphides. Hence we might conclude that these salts take part in the production of such highly sulphuretted animal substances as taurocholic acid, horny tissue, &c., but as substances which contain sulphur, such as legumin, gluten, &c., enter the animal body with the vegetable food, these highly sulphuretted substances, peculiar to the animal body, might also derive this element from

<sup>1</sup> Chem. Unters. über die Knochen u. Zähne. S. 226 u. 242.

<sup>2</sup> Lehrb. der Chem. Bd. 9, S. 695.

<sup>4</sup> Op. cit.

<sup>3</sup> Frauenmilch. S. 43.

<sup>5</sup> Jahresber. Bd. 16, S. 379.

the non-oxidized sulphur of the food. In the absence of any decisive experiments in favor of either of these views, we must for the present leave this question unanswered.

The experiments of Laveran and Millon<sup>1</sup> have shown that it is only when taken in large doses that the alkaline sulphates are carried off in the stools, small doses being absorbed in the intestinal canal and eliminated by the kidneys. We should, however, be in error, if we assumed, as Laveran and Millon seem to do, that this salt is simply absorbed in the intestinal canal; for it is well known that, after the use of alkaline sulphates, there is an excessive development of intestinal gas, which is especially rich in sulphuretted hydrogen.

This conversion of the sulphates into sulphides in the intestine during digestion is further established by the following facts. If I placed pure gluten, with milk-sugar and a little oil, in a dilute solution of sulphate of potash, and kept the mixture at a blood-heat, the mass first underwent the lactic fermentation, very soon became putrid, and in the course of 6 or 8 days, unmistakably developed sulphuretted hydrogen; in this way I was enabled, by the gradual addition of acetic acid, to remove the whole of the sulphuric acid from a mixture to which I had added 5 grammes of sulphate of potash. That the sulphate is, in like manner, deoxidized into the sulphide in the intestinal canal, where similar substances are brought in contact, is obvious from the composition of the stools which are discharged after the use of mineral waters, containing (like those of Marianbad) both sulphate of soda and carbonate of protoxide of iron.

In these fæces, which are usually green or black, I have recognized with certainty the presence of the sulphide of iron, but not of the bisulphide, as Kerston<sup>2</sup> seems to have done.

That the amount of sulphuric acid in the urine is chiefly due to the decomposition and oxidation of tissues containing sulphur is obvious from a comparison of the sulphates taken with the food and of those discharged by the urine.

As a mean of numerous experiments,<sup>3</sup> I found the sulphates discharged with the urine amounted daily to 7.026 grammes, while I was living on an ordinary mixed diet. After a strictly animal diet for 12 days, the sulphates rose to 10.399 grammes; and, after the use of a strictly vegetable diet, they fell to 5.846 grammes. During these experiments I drank nothing to allay my thirst but common spring water, which, besides a little gypsum, contained only small quantities of alkaline sulphates; so that the striking difference in the amount of the excreted sulphates could not be traced to that head. Moreover, the extraordinary augmentation of the urea in the urine excreted during my animal diet indicated that this corresponding augmentation of the sulphates depended on the same cause, namely, on a decomposition and oxidation of the substances taken as food.

<sup>1</sup> *Ann. d. Chim. et de Phys.* T. 12, p. 135.

<sup>2</sup> *Journ. f. Chirurgie von Walther und Ammon.* Bd. 2, S. 2.

<sup>3</sup> *Journ. f. pr. Chem.* Bd. 25, S. 2, and Bd. 27, S. 257.

## CARBONATE OF MAGNESIA.

This earthy salt occurs only sparingly in the animal organism. According to Berzelius,<sup>1</sup> it is not improbable that the magnesia in the bones is combined with carbonic, and not with phosphoric acid, and that the phosphate of magnesia found in the bones is only formed during the analysis. This view is supported by the circumstance that carbonate of magnesia is found with carbonate and phosphate of lime in many pathological concretions. If, however, the magnesia were combined with carbonic acid in the bones, it should be taken up with the carbonate of lime by dilute acetic acid, and neither in my experiments nor in those of von Bibra has this been the case.

Von Bibra<sup>2</sup> observes, in opposition to the view of Berzelius, that far more magnesia exists in the teeth than the carbonic acid found there can saturate.

Geiger<sup>3</sup> has published an analysis of a concretion extracted from the nose; it contained 76·7% of mineral substances, of which 8·3 were carbonate of magnesia. Bley<sup>4</sup> found 27·66% of carbonate of magnesia in a stony concretion from the peritoneum of a man.

A very large quantity of carbonate of magnesia exists in the urine of herbivorous animals, and hence we often meet with this salt in the urinary concretions of this class; it is very seldom found in human urinary calculi.

The urine of the ox, the camel, the horse, the rhinoceros, the elephant, the beaver, and the rabbit, deposits carbonate of magnesia with carbonate of lime. John<sup>5</sup> found 10% of carbonate of magnesia in the mucous deposit of the urine of a horse suffering from diabetes.

Lassaigne<sup>6</sup> found 4·8% of this salt, with carbonate of lime, in a calculus from the bladder of an ox, while Wurzer<sup>7</sup> obtained 4·06%, and Wackenroder<sup>8</sup> 3·522% of carbonate of magnesia from calculi obtained from the horse. A calculus from the bladder of a man, which was analyzed by Lindbergson,<sup>9</sup> contained, in addition to the phosphates of lime and magnesia, 2·55% of carbonate of magnesia, and only 3·14% of carbonate of lime. In two human calculi analyzed by Bley,<sup>10</sup> there were found 5·7% and 6·5% of carbonate of magnesia.

It is worthy of remark that, while plants, and especially the grasses, contain almost all their magnesia in combination with phosphoric acid, the urine of herbivorous animals so frequently contains carbonate of magnesia. We can hardly suppose that the phosphate of magnesia in the animal body, is robbed of its electro-negative constituent by a de-oxidation of the phosphoric acid, which is replaced by the weaker carbonic acid; it is much more probable that the combinations of lime with vegetable acids, conveyed into the animal body with the vegetable food, undergo such a decomposition with the phosphate of magnesia

<sup>1</sup> Lehrb. d. Chem. Bd. 9, S. 545.

<sup>2</sup> Op. cit. S. 94 and 287.

<sup>3</sup> Arch. der Pharm. Bd. 20, S. 212.

<sup>4</sup> Journ. de Chim. méd. 2 Sér. T. 4, p. 49.

<sup>5</sup> Ann. der Pharm. Bd. 18, S. 159.

<sup>6</sup> Buchner's Rept. 2. R. Bd. 2, S. 165.

<sup>7</sup> Mag. f. Pharm. Bd. 21, S. 247.

<sup>8</sup> Chem. Schriften. Bd. 6, S. 162.

<sup>9</sup> Schweig. Journ. Bd. 8, S. 65.

<sup>10</sup> Schweig. Journ. Bd. 32, S. 429.



either in the blood or in other parts, that bone-earth and a vegetable salt of magnesia are formed, the latter being subsequently converted into carbonate of magnesia. The fact that the urine of herbivorous animals is poor in phosphates, seems to confirm this view.

The egg-shell of birds contains not only carbonate of lime, but also carbonate of magnesia; both these salts are in part derived from the embryo during the incubation of the egg. (Prout<sup>1</sup> and Lassaigne.)<sup>2</sup>

### MANGANESE.

Minute quantities of this metal exist in the animal organism as elsewhere, in association with iron: manganese, however, seems to differ from iron, in being devoid of influence on the metamorphosis of the animal tissues, for it appears in comparatively larger quantities in the excretions, than in any of the fluids that take part in the vital functions. Like other heavy metals incidentally occurring in the organism, it is principally separated by the liver; hence it is found in comparatively large quantity in the bile.

Manganese has been found by Vauquelin<sup>3</sup> in the hair, and by Bley,<sup>4</sup> Wurzer,<sup>5</sup> and Bucholz,<sup>6</sup> in gall-stones and urinary calculi. Weidenbusch found 0.12% of proto-sesquioxide of manganese, and 0.23% of peroxide of iron in the ash of the bile, analyzed by Rose's method.

### ALUMINA.

This body never occurs in the animal organism; it has only been found in certain fossil bones into which it has undoubtedly entered by infiltration. Its absence in the animal organism is easily explained; any alumina conveyed into the intestinal canal enters into insoluble combination with organic substances, especially with the constituents of the bile, which cannot be resorbed.

After taking 3 grammes of basic sulphate of alumina within the space of 48 hours, I was unable to find a trace of alumina in the whole of the collected urine; it was, however, present in the ash of the solid excrements. The excrements were entirely devoid of odor, for some days after I took this substance.

### ARSENIC.

Devergie<sup>7</sup> and Orfila,<sup>8</sup> believed that they had found arsenic in all animal bones, and hence that it should be regarded as an integral constituent of the animal organism. Subsequent investigations have, however, shown that there must have been some fallacy in the method of analysis pursued by these chemists, and that this view is altogether erroneous.

When positive experiments seemed to show that arsenic existed in the

<sup>1</sup> Philosophical Transactions for 1822, p. 381.

<sup>2</sup> Journ. de Chim. Méd. T. 10, p. 193.

<sup>3</sup> Ann. de Chim. T. 58, p. 41.

<sup>4</sup> Op. cit.

<sup>5</sup> Op. cit.

<sup>6</sup> Op. cit.

<sup>7</sup> Ann. d'Hygiène publ. Oct. 1839, p. 482.

<sup>8</sup> Ibid. Juill. 1840, p. 163.

bones, chemists thought they had found an explanation of the apparent fact in the circumstance, that phosphorus and arsenic are so frequently associated together; if the discovery of Walchner and Schafhäütl that the sediments of most chalybeate waters contain arsenic had been then known, this would doubtless have been regarded as strong additional proof of the presence of arsenic in the animal organism.

Arsenic acts in so noxious a manner on the animal organism, even in the smallest doses (as we see from experiments on animals), that nature actively eliminates this deleterious substance as rapidly as possible from the body.

Meurer<sup>1</sup> has made experiments on horses (animals which, as is well known, can bear large doses of arsenic), and Von Bibra<sup>2</sup> on rabbits, from whence it appears that most of the arsenic is carried off with the solid excrements. Both observers also found the poison in the urine in no inconsiderable quantity. Of the solid parts of the animal body, the excreting organs, namely, the liver and kidneys, are those in which most arsenic is found; it has, however, also been detected in the heart, lungs, brain, and muscles. Some of these results are confirmed by the experiments of Duflos and Hirsch.<sup>3</sup>

Schnedermann and Knop<sup>4</sup> could detect no arsenic in the bones of a pig which had lived for three-quarters of a year in the neighborhood of the silver works at Andreasberg, where cattle and poultry do not thrive in consequence of the constant evolution of arsenical vapors.

### COPPER AND LEAD.

Both these metals have been found in very minute quantity in the healthy body by Devergie,<sup>5</sup> Lefortier,<sup>6</sup> Orfila,<sup>7</sup> Dechamps,<sup>8</sup> and Millon,<sup>9</sup> and were regarded by these chemists as integral constituents of all the soft parts, as well as of the blood; but it is only recently that any very decisive experiments on this subject have been instituted, and they, at all events, prove beyond a doubt, that copper exists in the *blood* of some of the lower animals, and in the *bile* of the ox and of man.

Millon believed that he had found them in the *blood*, but Melsens<sup>10</sup> has brought forward reasons, and even direct experiments against this view. Since, however, the presence of copper in the bile of man and the ox, has been determined with certainty, the blood must give traces of this metal, even though they be almost inappreciable. Moreover, E. Harless<sup>11</sup> has found copper in the blood, and more particularly in the liver, of some of the lower animals, namely, the *cephalopoda*, *ascidiæ*; and *mollusca*. This observer found copper in the liver of *Helix pomatia*; Von Bibra found it in the liver of *cancer pagyurus*, *acanthias*, *zeus*, &c.,

<sup>1</sup> Arch. d. Pharm. Bd. 26, S. 15.    <sup>2</sup> Untersuch. über die Knochen u. s. w. S. 112.

<sup>3</sup> Das Arsenik, seine Erkennung u. s. w. 1842.    <sup>4</sup> Journ. f. prak. Ch. Bd. 86, S. 471.

<sup>5</sup> Ann. d'Hygiène publ. Juill. 1840, p. 180.

<sup>6</sup> Ibid. p. 97.

<sup>7</sup> Mémoires de l'Acad. de Méd. T. 8, p. 522.

<sup>8</sup> Compt. rend. T. 27, p. 389.

<sup>9</sup> Journ. de Pharm. 3 Sér. T. 13, pp. 86-88 [also Compt. rend. T. 26, p. 41, and Ann. de Chim. et de Phys. 3 Sér. p. 372.—G. E. D.]

<sup>10</sup> Ann. de Chim. et de Phys. 3 Sér. T. 23, pp. 858-872.

<sup>11</sup> Müller's Arch. 1847, S. 148-157.

and observed that it stood in an inverse ratio to the iron. Copper was originally found in the bile and in gall-stones by Berzozzi,<sup>1</sup> and subsequently by Heller,<sup>2</sup> Görup-Besanez,<sup>3</sup> Bramson,<sup>4</sup> and Orfila.<sup>5</sup> I have been equally unsuccessful in demonstrating the presence of copper either in the human liver, or in the liver of the frog; in the latter case my experiment was made on 250 livers; and I have also failed in obtaining any indication of copper or lead in the blood, although I followed Milon's instructions.

There can be no doubt that the small quantities of copper which have been actually found in the fluids of the higher animals are only to be regarded as incidental constituents, while the experiments of Harless seem to indicate that in the lower animals the copper stands in an essential relation to the blood-corpuscles.

All the investigations which have hitherto been made, seem to indicate the liver as the organ in which deleterious substances, and especially those of a metallic nature, as, for instance, arsenic, lead, antimony, bismuth, &c., are accumulated, in order that they may be gradually eliminated with the bile. Hence, even if copper were constantly found in the blood or in the bile, it would afford no reason why we should regard this metal as an integral constituent of those fluids.

As copper has not only been found in many mineral waters (as, for instance, by Will,<sup>6</sup> Buchner,<sup>7</sup> Keller,<sup>8</sup> and Fischer,<sup>9</sup> but often in plants, and even in corn (Girardin),<sup>10</sup> there is no difficulty in accounting for its presence in small quantities in the organisms of the higher animals.

### SALTS OF AMMONIA.

Although many high authorities believe that they have found these salts in various parts of the animal body, yet if we put out of the question their occurrence in the excreted fluids, we must regard it as almost undoubted that no salt of ammonia is produced in the animal organism or found in the living parts.

In the sweat, especially in that from the axillæ, the occurrence of ammonia is incontestable. In the urine it is assumed to exist in larger quantities than is actually the case. In the solid excrements, which may be regarded as already in a state of decomposition, and which very soon develop ammonia when exposed to the atmosphere, Berzelius<sup>11</sup> believes that there is no carbonate of ammonia. Important as is the occurrence of ammonia in the vegetable juices for the renovation of the nitrogenous compounds, the animal organism appears to stand in little need of this substance. Indeed the process of decomposition by which the individual constituents of the organs are reduced to effete nitrogenous matter, by no means gives rise to the formation of ammonia, for in that case we

<sup>1</sup> Ann. di Chirurg. Milan, 1845, p. 32.

<sup>2</sup> Arch. f. Chem. u. Mikroskop. Bd. 3, S. 228.

<sup>3</sup> Unters. über Galle. Erlangen, 1848, S. 95.

<sup>4</sup> Zeitschr. f. rat. Med. Bd. 4, S. 193.

<sup>5</sup> Journ. de Chim. Méd. 3 Sér. T. 8, p. 484.

<sup>6</sup> Ann. d. Ch. u. Pharm. T. 55, p. 16.

<sup>7</sup> Jahrb. f. pr. Pharm. Bd. 15, S. 20-25.

<sup>8</sup> Journ. f. pr. Ch. Bd. 40, S. 442-447.

<sup>9</sup> Arch. der Pharm. Bd. 52, S. 268.

<sup>10</sup> Journ. de Chim. Méd. 3 Sér. T. 2, pp. 443-445.

<sup>11</sup> Lehrb. der Chem. Bd. 9, S. 180.

should certainly find a far larger quantity of the salts of this alkali in the excretions. Urea is the principal nitrogenous product of decomposition which is formed within the body from the nitrogenous substances.

The blood, chyle, lymph, and milk, the fluids of the egg, and the secretions of the serous membranes, either contain no ammonia or only extremely small quantities of it. In the pulmonary exhalation, on the other hand, small quantities of ammonia may always be recognized with great certainty.

Almost all histogenetic substances develop ammonia when treated with dilute acids or alkalies.

Observers have often believed that they had detected hydrochlorate of ammonia by the microscope after evaporating the alcoholic extract of animal fluids, when in reality, they saw the efflorescing forms of chloride of sodium, which, in the presence of certain organic matters (as, for instance, in the chyle) and especially when rapidly evaporated, separates in arborescent groups very similar to those of hydrochlorate of ammonia.

Lecanu and Denis failed in detecting any salts of ammonia in the blood; Marchand and Colberg were equally unsuccessful in reference to the lymph, and Schwartz and Simon, in reference to the milk.

Even in the urine the quantity of ammonia is extremely small, as is shown by the following experiments. I allowed the greater quantity of water in the morning urine to freeze, and thus obtained a very concentrated, almost wine-red urine, in which we might assume that there was no decomposition of the constituents; when carefully treated with caustic potash, it yielded a precipitate which even after remaining for a long time in contact with the urine, contained no uric acid; if salts of ammonia were contained in the urine, urate of ammonia would have been precipitated; but there was no deposit of this salt till after the addition of hydrochlorate of ammonia. Scherer and Liebig<sup>1</sup> have also convinced themselves of the absence of ammonia in normal urine. Heintz found that the ordinary urinary sediments consist of urate of soda with a little urate of lime, and only traces of urate of ammonia.

Boussingault<sup>2</sup> has recently attempted to determine the amount of ammonia in the urine by a new method, which depends upon the fact that all the ammonia may be developed from dissolved ammoniacal salts when they are evaporated to dryness in a vacuum with hydrated lime or carbonate of soda, at a temperature of from 40° to 50°, while the urea is not decomposed by such treatment. Proceeding in this way, Boussingault found 0.034% of ammonia in the urine of a child aged eight months, and 0.114% in that of a youth. It is, however, very questionable whether the nitrogenous matters of the urine, as, for instance, its colored extractive matters, which are decomposed far more readily than urea, may not, under these conditions, develop ammonia. At all events it would be remarkable if, after the use of the salts of ammonia, we were to find (as Bence Jones has done) not these salts, but their highest product of oxidation in the urine, while, when these salts have not been taken into the organism from without, the ammonia formed in the body

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 50, S. 198.

<sup>2</sup> Ann. de Chim. et de Phys. 3 Sér. T. 29, p. 472.

should not be decomposed, but should pass off as such in the urine. Moreover, it is not impossible that the ammonia found by Boussingault may have been formed within the body.

Marchand<sup>1</sup> was the first who ascertained with certainty that ammonia was present in the pulmonary exhalation; by means of the colorless hæmatoxylin discovered by Erdmann<sup>2</sup> he could detect it in the air of each individual respiration; moreover, when we employ sulphuric acid for the removal or determination of the water in experiments on the respiration, it is always found to contain ammonia.

In certain diseased conditions of the system very considerable quantities of ammonia are often found in the blood as well as in the urine. Winter<sup>3</sup> thought that the presence of ammonia in the blood explained the phenomena of typhus, but ammonia may be detected in the blood in all severe cases of acute disease, especially in variola and scarlatina; there is no more constancy in the presence of ammonia in the blood during typhus, than there is in the presence of the crystals of the triple phosphate in the excrements. It is by no means strange that in this state of the system the urine should contain ammonia; the urine is, however, richest in ammonia when it undergoes decomposition within the bladder, as in cases of inveterate vesical catarrh or diseases of the spinal cord.

### HYDROCYANIC ACID.

This acid never occurs preformed in the animal organism; even in the most varied of the metamorphoses and decompositions which occur during disease, we never meet with either the free acid or a metallic cyanide. This is readily accounted for, when we recollect that hydrocyanic acid, cyanogen, and the metallic cyanides, are only produced from nitrogenous substances at a high degree of temperature. But in spite of this, certain physiological chemists have shown no unwillingness either to assume that hydrocyanic acid, either in conjugation or in combination, exists preformed in histogenetic substances, or to avail themselves of its formation in the explanation of various chemico-vital processes; in short, to make it take a part in the equations by which they pretend to explain the different stages in the metamorphosis of the animal tissues. We only mention it here inasmuch as it belongs to the bodies which are produced during the artificial decomposition of animal substances, such, for instance, as acetic, valerianic, and cœnanthylic acids; we refer to the decomposition of hippuric acid by mere heat, and to the decomposition of histogenetic substances by bichromate of potash or binoxide of manganese and sulphuric acid.

### HYDROSULPHOCYANIC ACID.

This acid does not occur in a free state, but only as sulphocyanide of sodium [or potassium]. It was discovered by Treviranus in the *saliva*, and has as yet been found in no other fluid.

<sup>1</sup> Journ. f. pr. Ch. Bd. 33, S. 148, and Bd. 44, S. 35.

<sup>2</sup> Ibid. Bd. 27, S. 193-208.

<sup>3</sup> Ann. d. Ch. u Pharm. Bd. 48, S. 829.

Treviranus named it hæmatic acid (*Blutsäure*) ; and, because he found that it formed blood-red solutions with the persalts of iron, he attributed the color of the blood to sulphocyanide of iron.

For a very long time it has been disputed, whether the ingredient in the saliva, which gives rise to this red color with the persalts of iron, is actually sulphocyanogen. There is scarcely any subject in the whole domain of zoo-chemistry in which so many experiments have been made with such contradictory results. We believe, however, that no one who repeats the experiments of Pettenkofer<sup>1</sup> can entertain a doubt regarding the presence of sulphocyanogen in the saliva. Pettenkofer especially directs attention to two tests which he discovered for hydrosulphocyanic acid. Solutions of the acetate and formate of peroxide of iron are perfectly decolorized on boiling with alkaline chlorides, while this treatment has no apparent effect on sulphocyanide of iron : further, it is known that the persalts of iron do not decompose ferridcyanide of potassium ; but if we heat a solution of sulphocyanide of iron, hydrocyanic acid is developed, and there is a precipitate of Prussian blue. Pettenkofer applied this treatment to the alcoholic extract of the saliva, and thus ascertained the presence of sulphocyanogen. Other chemists had previously made use of a test that had been discovered for the sulphocyanides, namely, a mixture of two solutions of sulphate of protoxide of iron and sulphate of oxide of copper (when sub-sulphocyanide of copper is precipitated), with the view of detecting this substance in the saliva. The alcoholic extract of saliva is free from sulphuric acid (for the sulphates are insoluble in alcohol) ; hence Pettenkofer thought that he might make a quantitative determination of the sulphocyanogen in the saliva, by oxidizing the alcoholic extract with chlorate of potash and hydrochloric acid, and precipitating the sulphuric acid that was formed by chloride of barium.

Sulphocyanogen is almost always present in human saliva ; it is, however, occasionally absent, without any apparent physiological or pathological reason. It appears to be wanting in the secretion during salivation from any cause ; at least, I could never detect it during the ptyalism following the use of mercury or iodine, or occurring in the course of typhus or other diseases.

Sulphocyanogen occurs also in the saliva of the dog and the sheep ; I have examined the saliva of four different horses without detecting any traces of it ; Wright asserts, however, that it occurs in the saliva of that animal.

Considering the extremely small quantity in which it occurs, and that it is often absent without any apparent bad consequence, it seems hardly probable that the alkaline sulphocyanides take any definite part in the process of digestion.

I have noticed several healthy, vigorous young men, whose saliva contained no sulphocyanogen, and yet who enjoyed the best digestion.

It would be very easy to explain, by chemical formulæ, how sulphocyanogen might be formed from the histogenetic substances ; but, unfortunately, we as yet possess no facts to confirm us in the establishment

<sup>1</sup> Buchn. Repert. 2 R. Bd. 41, S. 289-313.

of any particular chemical equation ; it is better, therefore, frankly to confess that we know absolutely nothing regarding the place or the mode in which sulphocyanogen is formed in the animal organism.

## ANIMAL JUICES.

In our methodological introduction to physiological chemistry, the position was indicated which the theory of animal juices occupies, as an intermediate link between the theory of the organic substrata and that of the zoo-chemical processes; while the point of view was likewise defined from which this branch of physiological chemistry ought to be considered. It therefore only remains for us to make a few remarks on the mode of arrangement adopted in the following chapters, before proceeding to the special consideration of our subject. We purpose following in the main the same mode of arrangement which we endeavored to pursue in treating of the organic substrata; that is to say, we shall begin the notice of each object by considering its physical and chemical characters. The *mode of preparation* seems at first sight a matter of little moment, since these substances are usually submitted to examination in the condition in which they are directly yielded by nature. But although the exhibition of such objects does not require the aid of chemistry, we may often find it difficult to command the mechanical and physiological means requisite for procuring the substance in a pure condition, that is to say, unmixed and undecomposed. The result of the whole chemical operation is often dependent on the manner in which the object is exhibited, and it will be found that an unsuitable method of exhibition frequently leads to the adoption of wholly erroneous views in reference to the nature and function of an animal fluid. It is only when we are convinced that the animal fluid is presented to us in the same state in which it exists in the living body, that we can hope to obtain any physiological result from our investigation. As the exhibition of many animal fluids, moreover, frequently requires that the experimentalist should be familiar with certain anatomico-surgical operations, we think it will hardly be deemed superfluous, if we consider the methods adopted for procuring some of the animal juices.

When we have obtained a physiologically pure substance, studied its physical characters, and determined by the microscope the presence or absence of morphological elements, we must next consider the method in which the *chemical analysis* should be conducted. It is obvious that the plan and method of the analysis may vary in accordance with the special aims of the investigation, and will in general depend upon the nature of the constituents of the fluid. We have therefore thought it expedient to indicate the analytical methods of analysis suited to the different fluids. But as we are still on the very threshold of the inquiry, we can do no more than present the rudiments of a future organic analytical chemistry. As we have already observed, physiological chemistry, considered as an inductive science, requires most especially to be based



on exact principles amenable to calculation. Unfortunately, however, all chemists concur in admitting that a large number of analyses of the animal fluids rank among the most slovenly and unprincipled investigations of their science. How many of these show at the first glance that they are utterly worthless! In such a state of things it will scarcely be deemed superfluous to specify some few of the properties which it is necessary for the analysis to possess in order to render it of any value in a scientific point of view.

As we have already referred to the qualitative and quantitative determinations of the individual animal substrata, it only remains for us to observe, in reference to compound fluids generally, that in the *qualitative* analysis of the animal juices, the largest possible quantity should be employed—a point the importance of which was first fully demonstrated by Liebig's investigations regarding the juices of the flesh, &c.\* For *quantitative* zoo-chemical analyses, however, the converse rule holds good, more especially in analyses of the blood. We generally find that too large a quantity has been employed for the determination of the individual constituents in the majority of the blood-analyses on record. As a general rule, it may be asserted that quantitative analyses of the blood and of similar fluids, are the less exact in proportion to the quantity of the substance analyzed. This depends partly on the difficulty frequently experienced in passing animal fluids, even when diluted, through a filter, partly on the readiness with which many of the constituents are decomposed, but principally on the impossibility of perfectly and uniformly drying large quantities. In order to avoid as much as possible these and other impediments, we must only employ small quantities of the object in our analysis.

Notwithstanding every care and precaution, difficulties will however present themselves in the analysis of animal substances, which may escape even the closest attention; and hence, more than in the analysis of any other substances, it is absolutely necessary to institute a rigid *controlling comparison* of the various results, and even a partial repetition of them. Since the same quantity of an animal fluid serves for the determination of only a few of its constituents, our means of controlling the analyses are increased in proportion to the number of determinations made independently of each other. Thus, for instance, in seeking to ascertain the amount of coagulable matters contained in a fluid, the analysis should always be controlled by extracting the solid residue of the fluid with alcohol, ether, and water, and then comparing the quantity of the insoluble matters with the number representing the protein-body determined by coagulation. Thus too, ash-analyses should always be controlled by comparing the mineral constituents of the individual extracts with those of the collective ash. A perfect coincidence would certainly prove the inaccuracy of the analysis in both these cases; for the coagulated substance, unless it has been expressly deprived of its fat, still contains fat, and sometimes even other substances soluble in alcohol but not in water, and which, on the other hand, cannot occur in that portion of the solid residue of the fluid extracted with alcohol and ether, and insoluble in water; for this must always contain more earthy salts than the albumen that has been completely coagulated by the addi-

tion of a weak acid. In like manner the analysis of the collective ash cannot coincide with that of the individual extracts, since, for instance, the sulphur contained in the coagulable matters is unable to exert a metamorphic action on the soluble salts of the extracts, whilst the composition of the combined ash must be altogether different, partly on account of the sulphuric acid formed from the unoxidized sulphur of the protein-bodies, partly from the difficult combustion of albuminous substances, and partly from other causes. Although this method may not afford any strict means of controlling these relations, it furnishes a more correct view of the true nature of the substances dissolved in the fluid. This is, to a certain degree, a physiological check; but it would be superfluous to indicate any special methods of control, since every analysis of an animal fluid, and even almost every individual determination, admits of being tested by the most rigid purely chemical checks. We shall find that these controlling or controlled analyses frequently afford the most unexpected proofs of the value of the method of analysis, by the light they throw upon the true constitution of the object under examination.

C. Schmidt<sup>1</sup> has employed an ingenious and original method for ascertaining the correctness of the complete analysis of an animal fluid; it consists in comparing the empirically found specific weight, with the sum of the specific weights of the individual constituent parts, according to the proportions yielded by the analysis. Such a controlling calculation of the density cannot of course be based upon the specific weights of the dried substances and of the water; since all substances when dissolved in water undergo a condensation with it. It is a highly important circumstance in relation to the complete physiological consideration of the animal fluids and of the mechanical metamorphosis of matter, that the dissolved substances do not occur, as has been generally supposed, in a mere condition of mechanical distribution and admixture, but that when dissolved in different quantities of water, they enter into different hydrate-like combinations with that fluid, and consequently exhibit various degrees of condensation. Schmidt has determined the coefficients of condensation for the ordinary constituents of animal fluids, and made the density of the solutions which contain exactly 10% of solid substances (at  $+15^{\circ}$  C. in vacuo) the basis of his controlling check. Knowing the relations of the individual constituents from the analysis, we may easily calculate the specific gravity of the collective fluid from the sum of the coefficients of condensation, and may then compare it with the empirically found specific gravity.

The following illustration will serve to elucidate the table compiled by Schmidt for the purpose already stated. The specific gravity of chloride of sodium (at  $+15^{\circ}$  C. in vacuo) is  $= 2.1481$ ; hence, if the salt in a solution containing 10% of chloride of sodium were distributed in the water without condensation, this solution would have a specific gravity of 1.0565; for 10 parts of dry chloride of sodium (its specific gravity being  $= 2.1481$ ) occupy the space of 4.655 parts of water; with 90 parts of water, therefore, the solution should occupy the space of 94.655 parts of water; but when we examine the specific gravity of

<sup>1</sup> *Charakteristik der Cholera.* Mitau, 1850, S. 22–28.

such a solution, we find that it is higher, that is to say = 1.0726. In accordance with this density, ten parts of chloride of sodium and ninety parts of water, occupy only the space of 93.231 parts of water; hence a condensation of 1.424 parts must have taken place; for every 100 volumes there would, therefore, be a condensation of 1.505 volumes between one part of chloride of sodium and nine parts of water. With these preliminary remarks we proceed to give Schmidt's table.

SUBSTANCE.	Density of the solution containing 10g of solid substance.	Density of the dry substance.	Percentage, according to volume, of the condensation occurring in the solution containing 10g.
Chloride of sodium, . . . . .	1.0726	2.1481	1.505
Chloride of potassium, . . . . .	1.0653	1.9787	1.348
Sulphate of potash, . . . . .	1.0833	2.6616	1.541
Phosphate of potash, $2\text{K.O.P.O}_5$ , . . . . .	1.0960	2.4770	2.974
Phosphate of soda, $2\text{Na.O.P.O}_5$ , . . . . .	1.0994	2.3735	3.455
Potash, . . . . .	1.1001	2.6560	3.035
Soda, . . . . .	1.1484	2.8050	6.933
Phosphate of lime, $3\text{Ca.O.P.O}_5$ , . . . . .	1.0807	3.0976	0.744
Phosphate of lime, $2\text{Ca.O.P.O}_5$ , . . . . .	1.0896	3.0596	1.600
Phosphate of magnesia, $2\text{Mg.O.P.O}_5$ , . . . . .	1.0913	3.0383	1.776
Phosphate of iron, $\text{Fe}_2\text{O}_3\text{P.O}_5$ , . . . . .	1.0880	3.0661	1.447
Urea, . . . . .	1.0275	1.8369	0.160
Glucose, $\text{C}_{12}\text{H}_{22}\text{O}_{12}$ , . . . . .	1.0396	1.3860	0.766
Fibrin, . . . . .	1.0270	1.2858	0.420
Albumen, . . . . .	1.0268	1.2746	0.426

We forbear offering any remarks in this place on the interesting points of view which are opened to us by Schmidt's admirable investigation, as we must return to the full consideration of this subject when we treat of the mechanical metamorphosis of tissue.

Schmidt employs specific gravities as a check on analyses, in the following manner. The analysis of a specimen of serum, whose specific gravity was found to be 1.0292, gave 82.59 p. m. of organic constituents, 0.283 p. m. of sulphate of potash, 0.362 of chloride of potassium, 5.591 of chloride of sodium, 0.273 of phosphate of soda, 1.545 of soda, 0.300 of phosphate of lime, and 0.220 of phosphate of magnesia. The following is the manner in which the check is applied :

			Water.
82.586 grammes of albumen with extractive matters (the condensation dependent on solution being allowed for) fill the space of			61.471
2.836 " of the 10% solution of sulphate of potash,	"	"	2.618
3.616 " " chloride of potassium,	"	"	3.395
55.914 " " chloride of sodium,	"	"	52.129
2.726 " " phosphate of soda,	"	"	2.480
15.454 " " soda,	"	"	13.457
2.997 " " phosphate of lime,	"	"	2.773
2.197 " " phosphate of magnesia,	"	"	2.013
168.326 grammes of the collective solution,		"	140.336

Hence we calculate the density of serum, having this composition, to be 1.0288; for  $140.336 : 168.326 :: 1 : x (=1.0288)$ .

After having acquainted ourselves with the chemical constitution of

the animal juices in their normal state, we have next to consider the modifications experienced by the fluids in question under *different physiological and pathological relations*, considering at the same time the composition of the corresponding juices in different classes of animals. The latter indeed constitutes the most common subject of our physiologico-chemical inquiries, and the main basis of our investigations.

Before we consider the *pathological* relations, it will be necessary to make a few preliminary remarks. What we have already said in reference to the mode of treating pathological chemistry, sufficiently demonstrates how visionary are all anticipations of the formation of a perfect humoral pathology, which indeed is a science that has no existence except in the dreams of mere enthusiasts. According to the principles on which we would base our consideration of pathological processes, we cannot group the physical and chemical alterations observed in animal juices within the generally recognized classes of disease, but must arrange them in harmony with the internal, that is to say, the chemical constitution of the pathological objects. It seems to us, that we should be assuming an entirely false point of view, were we to start from conventionally named diseases, as tuberculosis, carcinoma, &c. Notwithstanding the frequent objections advanced against the ontological modes of definition in use for diseases, an entirely specific character has nevertheless been ascribed to these hackneyed designations of certain forms of disease, since otherwise the idea that tubercles are mere depositions of exudation, and similarly erroneous notions, could never have become current. That we may avoid such a fictitious species of physiology, we shall adhere as strictly as possible to the object itself, merely reverting, where it is absolutely necessary, to its conventional predicate.

Although we may describe the bile in the dead body as poor in solid constituents after violent inflammations, as still thinner and more watery in typhus, and sometimes deficient, and at other times abundant in solid constituents in tuberculosis, we must, nevertheless, consider this designation of the conditions in which the bile is found to be more concentrated or more attenuated, as wholly irrational; for we ought simply to have said that in those conditions in which, to borrow an expression from pathological anatomy, the morbid process had localized itself, or, in other words, where the blood had lost some of its solid contents, in consequence of extensive exudations or other considerable losses of the juices, this property of the blood was reflected as it were in the secretions and excretions, and a less consistent and poorer bile was secreted; whilst in those cases in which the blood is found to be denser and to contain more solid constituents, as for instance in cholera, the bile in the body after death is viscid and deficient in water.

Another point of physiological importance in relation to the animal fluids, is the investigation of the quantities in which they are *formed or secreted*, and this is far more necessary than we should be disposed at first sight to infer. We have already shown, in our methodological introduction, the importance to be attached to the statistical method of examining the metamorphosis of matter, and recognized it as one of the most valuable aids in physiologico-chemical investigation, for although it still leaves us in the dark as to the nature and objects of such a process,

it defines certain boundaries beyond which we cannot strain our interpretation of animal phenomena, or extend our experiments, without falling into the most obvious errors. Such a limitation of hypotheses is above all most necessary in a science which may still be said to be in its infancy. The benefits derived from this statistical method are not, however, merely negative; for it affords the surest basis for the recognition of that branch of physiological chemistry which promises to yield the richest fruits to future inquirers. The most direct and attainable aim of our investigations must be the elucidation of the quantitative relations of the interchange of the different animal substances through the different organs, tissues, closed and open cavities, and, finally, the external world. In the present low state of our knowledge of the chemical substrata of the human organism in health as well as in disease, it is to a development of the mechanical metamorphosis of matter based upon physical laws, and referable to simple numerical calculations, that we must look for the most brilliant results. The groundless hypothesis of crases and dyscrases stimulated zeal for the chemical investigation of morbid products; but what have we learnt from the innumerable analyses of morbid blood and urine, beyond the fact that the quantitative proportions of the ordinary constituents of these juices have undergone certain modifications? As we have but little chance of success, at least for the present, in seeking for deleterious matters, specific contagia, a *materia peccans*, &c., we should rather direct our inquiries to the elucidation of the quantitative relations of the substances known to us, and to their distribution in the different animal juices. But a meagre enumeration of the barren results of chemical analyses in percentage tables is insufficient for this end, since what we require is to bring these results into harmony with the relations of mass existing between the different animal fluids, and with the amount of motion occurring between the juices that are separated by membranes and cells. If, for instance, we compare the quantities of the substances occurring in the secretions during disease with those which remain in the blood, we shall arrive at results yielding the most interesting materials to physiological mechanics, in elucidating the course of morbid processes and the causal connection existing between entire groups of symptoms, as has been ably shown by C. Schmidt in his admirable investigations on the processes of transudation in cholera, Bright's disease, dysentery, and dropsical conditions. The knowledge of the quantitative relations in which each animal fluid and its individual constituents are formed or secreted, supplies the basis of the statistics of animal molecular motion; we purpose, therefore, entering into a special consideration of the mechanical metamorphosis of matter in the animal organism, in our second volume, where we shall further endeavor to reconcile the results of the quantitative physiologico-chemical inquiries with the theories of the imbibition of animal membranes, of endosmosis, and of the transudation depending upon the elasticity and thickness of the membranes as well as upon the rapidity of the motion of the blood. Without such points of support, based on physical laws and arithmetical conclusions, few hypotheses regarding nutrition and secretion, and the metamorphosis of the body generally, can attain any degree of logical accuracy. We have therefore regarded it as perfectly falling within the

province of physiological chemistry to give the quantity of the matters secreted, and the amount of chemical motion of each animal juice, as far as the state of science enables us to form such estimates.

A larger portion of the systematic treatment of the animal juices will be devoted to a consideration of the *metamorphoses* experienced by each separate object within the living animal organism, and the changes and decompositions observed in the same substance external to the sphere of vitality. If we subjoin, as in our notice of the animal substrata, those circumstantial data which can alone justify us in considering the *genetic* development of each object, we shall be in possession of all the elements, as far as the present state of science permits, for forming an opinion regarding the *function* or *physiological value* of every individual animal fluid. By such a course, we shall certainly be carried within the province of physiological processes; but in considering the metamorphosis of animal matter generally, and the processes of digestion, respiration, and nutrition, in their systematic connection, our views of the chemistry of the animal body should not be diverted to individual points, but rather be made to combine with the conclusions obtained by simple induction, in reference to the function of the individual chemical agents.

If ever we cherished the hope of combining the results of former inquiries into one scientific whole, constituting a purely inductive branch of science, in accordance with our view of the method in which physiological chemistry, and more especially the theory of the animal juices, should be treated, our courage would fail, as indeed it often has done, when we attempted the accomplishment of such a task. We believe that we have already sufficiently explained our view of the very great deficiency of our knowledge in this department of the physical sciences; but here it is less a want of positive knowledge than a redundancy of materials that renders it a matter of almost insurmountable difficulty to demonstrate with clearness the pure and unadulterated character of science free from pretentious delusions. A cursory glance at the confused mass of materials accumulated before us, presents a view of disorder requiring Herculean efforts to disentangle them. We confess that we have therefore abstained from attempting in the following pages to give the whole mass of the results that have been obtained within this department of science from all experiments and observations, whether good or bad; limiting ourselves to facts collected by the best observers, which, as far as our powers and experience permitted, we have compared with the results of our own observations, testing the different conclusions and hypotheses by a course of logical inquiry. Without reference to the present work, which we have designated as a mere attempt, in genuine sincerity, and apart from all pretended modesty, it will not be denied that far greater service will be done to the theory of animal juices as well as to physiological chemistry, by an experimental criticism, than by the most careful collection of all that bears upon the subject in literature. In our attempt to sift the rich mass of materials presented to our notice, we shall endeavor to abstain from all mere polemical criticism, and adhere to facts only, which must ever constitute the only solid support of natural science.

## SALIVA.

The saliva discharged from the mouth is not merely a mixture of the fluids secreted by the different salivary glands, but also contains, as an essential constituent, the buccal mucus, or the secretion of the mucous membrane of the oral cavity. The mixed or ordinary saliva is therefore by no means identical with the secretions of the different salivary glands, from which it differs both in its chemical characters and in its physiological action.

The *mixed saliva* of man and of most of the mammalia exhibits the following properties: it occurs as a rather turbid, opalescent, or faintly bluish-white fluid, which is somewhat viscid and capable of being drawn out in threads, and is devoid of odor and taste. After standing for some time, it deposits a mucous grayish-white sediment, which, when examined under the microscope, is found to consist chiefly of pavement epithelium, often united so as to form shreds, and what are termed mucus-corpuscles, which are usually a little larger than pus-corpuscles, and generally exhibit a large, lenticular, excentric nucleus, even without the application of any special reagents. The specific gravity of mixed saliva is liable to variations even in the normal state; for its density is partly dependent on the quantity of mucus that may be mixed with it, and partly on the greater or less attenuation of the glandular secretion; its usual variations in man are between 1.004 and 1.006; it may, however, in the normal state rise to 1.008 or 1.009, or, on the other hand, it may sink to 1.002. Normal saliva presents a more or less distinctly alkaline reaction: it has no poisonous effect either on plants or animals.

There is scarcely any animal fluid in which it is of such importance that the specimen we are examining should be perfectly fresh; for none becomes so rapidly changed and so soon completely decomposed as the saliva. A disregard of this fact is the cause of many of the errors which have led to the most remarkable views regarding the saliva. It is thus, probably, for instance, that Wright<sup>1</sup> has ascribed to this secretion many properties which have either not been at all noticed by other observers, or at all events in a less degree. We may refer, by way of example, to the taste of the saliva, which, according to Wright, is "sharp, saline, and slightly astringent,"—a statement which I must agree with Jacobowitsch<sup>2</sup> in totally denying; for, in opposition to Wright's assertion, I have always found the saliva of healthy persons to be tasteless. The injurious action of saliva on vegetable and animal organisms, which has been observed and described by Wright, depends for the most part, as may be shown by positive experiments, on the fact of its not being perfectly fresh.

The morphological elements of the saliva owe their origin to the mucous membrane of the buccal cavity, and in a lesser degree also to that of the salivary ducts; hence, these bodies will be described in the chapter on "Mucus." In examining expectorated saliva we sometimes find not only epithelium and mucus-corpuscles, but also fat-globules, and occa-

<sup>1</sup> On the Physiology and Pathology of the Saliva. *Lancet*, 1842.

<sup>2</sup> De Saliva, diss. inaug. Dorpati, Liv. 1848, p. 12.

sionally the remains of food, as, for instance, vegetable cells or beautifully macerated muscular fibre, and still more rarely vibriones, which take their origin in mucus or fragments of food retained for a long time between the teeth, or in hollow carious teeth.

The presence of *mucus-corpuscles* in normal saliva or the buccal mucus has been called in question, and it has been asserted that they only occur after some slight irritation of the mucous membrane of the mouth, as, for instance, after smoking tobacco; but I have always been able to detect some of them in the buccal mucus of healthy persons (even of such as are not in the habit of smoking); and as they likewise occur in the saliva of animals, as for instance, of dogs and horses (Maggendie,<sup>1</sup> Jacobowitsch),<sup>2</sup> it cannot be doubted that the buccal mucous membrane, even in its perfectly normal state, throws off these mucus-corpuscles with the epithelial plates, the former indeed being nothing more than abortive epithelial cells.

The extreme variability of the *specific gravity* of the saliva of even the same person, under different physiological relations, may be easily demonstrated by a few experiments. I made some experiments in reference to this point with the parotid secretion of a horse, in whom an artificial salivary fistula was established, by exposing and opening the duct of Steno. Very shortly after the operation the parotid saliva had a density of 1.0061; ten minutes afterwards, when the animal had drank about six pounds of water, and had eaten some hay, the density sank to 1.0051; after having nothing to drink for twelve hours, a feed of hay caused a free secretion of saliva, whose specific gravity was as high as 1.0074. Wright has pointed out that human saliva is denser after food has been taken than in a fasting condition. He found that the saliva of a healthy man who had lived for a week on a mixed diet, varied in its density from 1.0079 to 1.0085; while, after a purely animal diet for an equal time, it varied from 1.0098 to 1.0176; and, after a purely vegetable diet, from 1.0039 to 1.0047. According to this author, moral emotions, atmospheric changes, light, sound, &c., exert an influence on the density of the saliva. From numerous experiments made on 200 healthy persons, he found that the specific gravity of the saliva varied between 1.0089 and 1.0069, a result which is far higher than I have obtained from my experiments. It is possible that the more abundant use of an animal diet amongst the English may have caused the higher density which was found by Wright.

In relation to the *alkalinity* of the saliva, any one may readily observe for himself that, during and after eating, the alkaline reaction increases, while during fasting it very much diminishes, or altogether disappears; indeed, in some persons who are apparently healthy, the saliva during fasting has a weak acid reaction, although immediately after the use of solid food it again becomes alkaline (Hünefeld,<sup>3</sup> Mitscherlich,<sup>4</sup> Wright, Jacobowitsch). According to Wright, the quantity of soda in the saliva of healthy men varies between 0.095 and 0.3538; in that of dogs between 0.151 and 0.6538; in that of sheep between 0.087 and 0.2618; and in that of horses between 0.098 and 0.5138.

<sup>1</sup> Compt. rend. T. 21, p. 905.

<sup>2</sup> Chemie und Medicin. Berlin, 1841, S. 48-60.

<sup>3</sup> Op. cit. p. 16.

<sup>4</sup> Pogg. Ann. Bd. 27, S. 320-347.



We only record these numbers in order to give an approximate measure of the quantity of acid which may be saturated by the saliva; for these numbers are calculated for soda, while in the saliva of graminivorous animals there is often much potash, and always a large quantity of lime, which is expelled from its combination with non-acid organic substances by the very weakest acids, and as for instance even carbonic acid. Frerichs<sup>1</sup> found that 100 grammes of saliva, secreted by a man smoking tobacco, were neutralized by 0.150 of a gramme of sulphuric acid.

According to Wright, the quantity of alkali in the saliva is increased after the use of fatty, aromatic, acid, spirituous, and more particularly indigestible kinds of food and drink.

In attempting to collect *pure* human saliva, we must avoid the use of irritants—such as tobacco, either smoked or chewed, or aromatics—which, although they increase the secretion, become mixed with it, and render it comparatively unfit for examination. The simplest method of collecting a large quantity of saliva in a short time, is by exerting a strong pressure under the chin, and at the same time tickling the palate with a feather; a feeling of strangulation rapidly ensues, during which the saliva is ejected from the mouth. The best method of collecting the saliva of animals, is by presenting them with their favorite food after they have been kept for some time fasting; the secretion flows freely on pressing the nostrils in a backward direction.

The method which Magendie and Lassaigne adopted for the purpose of collecting the mixed saliva of animals, namely, cutting into the œsophagus, cannot be avoided for certain experiments, but, for ordinary purposes, it is not only cruel and indirect, but also unphysiological; for how can we expect, that after such an inroad on animal life as must arise from the exposure and opening of the œsophagus, a secretion can remain in its normal state?

We have already mentioned that ordinary saliva is a mixture of the secretion of the buccal mucous membrane and of several glands; we now proceed to notice these secretions individually.

PAROTID SALIVA has hitherto only been accurately examined in man by Mitscherlich<sup>2</sup> and Van Setten;<sup>3</sup> the parotid secretion of horses and dogs has, however, very often been analyzed. It is usually perfectly limpid and colorless, devoid of smell and taste, incapable of being drawn out in threads, and of a distinctly alkaline reaction. In a male patient, Mitscherlich found that the specific gravity varied from 1.0061 to 1.0088; in dogs it was found by Jacobowitsch to vary from 1.0040 to 1.0047; and in horses I found it to range from 1.0051 to 1.0074.

The observations of Mitscherlich on the parotid saliva of a man suffering from chronic disease, show that prolonged hunger or the use of indigestible and stimulating food, causes the secretion of a concentrated saliva. Moreover, this observer found that in the fasting state it was always acid, and that it was only alkaline during meals. Magendie and Rayer perceived a gradual diminution in the specific gravity of the parotid saliva of a horse in whose Stenonian ducts fistulous openings had been established.

<sup>1</sup> Wagner's Handwörterb. d. Physiol. Bd. 3, Abt. 1, S. 760.

<sup>2</sup> Op. cit.

<sup>3</sup> De Saliva ejusque vi ut utilitate. Groning. 1837.

In regard to the chemical constituents of the parotid saliva, it may be observed that the results of those who have experimented on the subject, do not altogether coincide, probably from their having operated on the saliva of different classes of animals. The following may, however, be regarded as constant ingredients of this secretion :

(a) *Potash, soda, and lime*, combined with an *organic matter*: this compound is one of the most important of the constituents of the saliva, being that on which several of the properties of this fluid are dependent; it is similar to, but not identical with, albuminate of soda, and corresponds in part to the ptyalin of Berzelius and others.

Magendie, Jacubowitsch, and others, assume that alkaline carbonates are present in the saliva, but their quantity must be extremely small in the fresh secretion; the alkaline carbonates are produced during the different steps of the chemical analysis, by the access of atmospheric air. The formation of carbonate of lime is extremely evident when the parotid secretion of the horse is exposed to the air, for, like lime-water, it attracts carbonic acid, and most beautiful microscopic crystals of carbonate of lime are deposited. The organic matter, the ptyalin, is difficult of solution, although not absolutely insoluble, in water, after its separation from the alkalies or from lime, by carbonic or some other acid; hence human saliva, and that of the dog, is sometimes rendered turbid, and is sometimes apparently unaffected by acids; the precipitate consists of amorphous flocculi, which are difficult of solution in pure water, but dissolve readily if an alkali or an acid be added. We find this substance in part still combined with an alkali, both in the aqueous and in the spirituous extracts; it may be obtained in the greatest purity from the latter, after extraction with alcohol and ether. It then forms an almost gelatinous, colorless substance, which is more or less insoluble in water, according as the alkali has been more or less thoroughly removed by carbonic acid, or some other means. The alkaline solution of this substance yields, on the addition of a little acetic or nitric acid, a flocculent precipitate, which dissolves freely in an excess of acetic acid; when boiled with hydrochlorate of ammonia, or with sulphate of magnesia, the alkaline solution of ptyalin becomes strongly turbid. The alkaline (but not the neutralized) solution of this substance is precipitated by tannic acid, corrosive sublimate, and basic acetate of lead, but not by alum or sulphate of copper. The acetic acid solution becomes strongly turbid on the addition of ferrocyanide of potassium; when boiled with nitric acid, this substance forms a yellow solution. In these respects, it is very similar to albuminate of soda, and to casein, but it must by no means be confounded with them. I have principally studied the properties of this substance in cases in which it had been obtained from the parotid saliva of the horse, and I have arrived at the conclusion, that the differences observed by Berzelius, Gmelin, and others, in reference to ptyalin, may be easily explained. In no other animal fluids could I recognize a substance perfectly identical with this ptyalin.

It is singular that Magendie, in his investigations regarding the parotid saliva, has overlooked the circumstance that it abounds in lime (and assumes only the presence of bicarbonate of potash); while Jacubowitsch<sup>1</sup>

<sup>1</sup> Op. cit. p. 20-22.

constantly found carbonate of lime in the parotid saliva of dogs. It is possible that differences of food may exert the same influence on the saliva as they do on the urine of horses; for, as we shall subsequently show, the urine of these animals sometimes abounds in carbonate of potash, and sometimes in carbonate of lime; but whenever I analyzed the saliva of the horse, I always found it very rich in lime.

(b) An *extractive matter* soluble in *alcohol* and in *water*, which is precipitable by tannic acid, but not by alum.

(c) *Sulphocyanide of potassium*, whose presence has been detected by Mitscherlich, Jacobowitsch, and Gmelin, in the parotid saliva of man, the dog, the horse, and the sheep.

I have not observed any reddening on the addition of perchloride of iron to the parotid secretion of the horse.

(d) The *potash-salt* of a not very volatile acid belonging to the *butyric acid* group (caproic acid?); it crystallizes in a beautiful efflorescent form, which, under the microscope, resembles the tufts of margaric acid.

(e) A little *epithelium*, and some scattered *mucus-corpuscles*.

(f) The *chlorides of sodium* and *potassium*.

(g) *Phosphates*, in very small quantities.

(h) A trace of *alkaline sulphates*.

The following statements may suffice in reference to the *quantitative* relations of the constituents of the parotid secretion: in the parotid saliva of man, Mitscherlich found from 1.468 to 1.632%, and Van Setten 1.62% of *solid constituents*; in that of a dog, Jacobowitsch found only 0.47%, while Gmelin found 2.58%; in that of the horse Magendie found an average of 1.1%; while as the mean of six experiments with different specimens of saliva, I determined the solid constituents at 0.708%.

In the human secretion, Mitscherlich found nearly 0.525% of *ptyalin* associated with an alkali; in that of the horse, I found on an average 0.140% of pure ptyalin (after the extraction of the mineral substances contained in it).

The alkaline ptyalin obtained from the water-extract and from the spirit-extract insoluble in alcohol, constituted 23.332% of the solid constituents of the saliva of the horse, and yielded 5.675% of ash, which consisted almost entirely of alkaline carbonates and lime.

The *alcohol-extract* of the secretion amounted in man, according to Mitscherlich, to about 0.1%; in that of the horse, I found it amount to 0.0988%.

The alcohol-extract of the saliva of the horse amounted, according to the mean of my experiments, to 13.936% of the solid residue, and yielded 3.812% of ash, consisting chiefly of alkaline chlorides.

No quantitative determination of the *sulphocyanide of potassium* contained in the parotid saliva has yet been attempted.

In the parotid saliva of the horse, I found 0.0403% of a compound of a *fatty acid* and *potash*.

The ether-extract amounted to 5.703% of the solid residue, and contained 1.102 parts of potash (as determined by bichloride of platinum from the ash).

\* The *insoluble matter* removed by filtration, and consisting of epithelium with salts, amounted according to Mitscherlich, to 0.005%, while I found it as high as 0.124% in the saliva of the horse.

The insoluble portion of the saliva of the horse consisted, for the most part, of carbonate of lime; after its abstraction, and that of the ash generally, the insoluble organic matter was very minute; the solid residue contained 17·55% of insoluble matters, in which were 13·453 parts of ash; hence the epithelium amounted to only 4·097% of the whole solid residue.

According to the determinations of Mitscherlich, the solid residue of the parotid saliva in man contains about 45·7% of mineral constituents, in which there are contained 35·4 parts of chloride of potassium, and about the same quantity of potash and soda (after deducting the carbonic acid); in this secretion from the dog, Jacobowitsch found that the ratio of the organic to the inorganic matter was as 29·8 : 70·2; the latter consisted of 44·7 parts of alkaline chlorides, and 25·5 of carbonate of lime. In 100 parts of the solid residue of the parotid saliva of the horse, I found 53·9 parts of ash, in which there were 21·764 parts of chloride of potassium, 16·983 of carbonate of potash, and 11·226 of carbonate of lime, while there were only 0·882 parts of the phosphates of lime and magnesia, 0·805 of the sulphate, and 2·240 of the phosphate of soda.

THE SECRETION OF THE SUBMAXILLARY GLANDS of dogs has been carefully examined by Bernard<sup>1</sup> and Jacobowitsch; it is, like the parotid saliva, a colorless, limpid, tasteless, and inodorous fluid, and is devoid of all morphological elements. Jacobowitsch determined its specific gravity at 1·0041; the reaction is less strongly alkaline than that of the parotid saliva; it contains far less lime in combination with organic matter, and therefore attracts less carbonic acid from the air than the previously described secretion; in other respects, it contains precisely the same constituents, including the sulphocyanide of potassium. Bernard regards the viscosity of this secretion as constituting an essential difference between it and the parotid saliva, and Jacobowitsch likewise noticed this peculiarity in the submaxillary fluid. According to the last-named observer, it yielded 0·855% of solid residue, in which the ash amounted to 0·566 parts; so that here the ratio of the organic to the mineral constituents was as 33·8 : 66·2; the latter contained 52·6 parts of alkaline chlorides and 13·6 of carbonate and phosphate of lime and magnesia.

Bernard directs attention to the circumstance that an infusion of the parotid gland is very aqueous, and cannot be drawn out in threads, while an infusion of a piece of the submaxillary gland is as viscid as the secretion collected from Wharton's duct.

THE SECRETION OF THE BUCCAL MUCOUS MEMBRANE in dogs has been examined by Jacobowitsch; but the secretions of the orbital and of the sublingual glands (which latter are, however, very little developed in dogs) were mixed with it. This fluid was very viscid and tenacious, frothy and colorless, but extremely turbid from the presence of an enormous number of epithelial cells, which were not deposited when the fluid was allowed to stand. The fluid had an alkaline reaction, and did not coagulate on heating; it left 0·999% of solid residue, in which were 0·385 parts of organic and 0·614 of inorganic matter. The insoluble salts contained no carbonate of lime.

<sup>1</sup> Arch. gén. de Méd. 4 Sér. T. 13, p. 1-29.

Jacobowitsch collected the mucous secretion of the mouth by tying Steno's and Wharton's ducts, keeping the animal's nostrils open, and retaining the head in an inclined position, so that the animal being unable to swallow, the mucus flowed from the mouth. He collected the secretions of the parotid and submaxillary glands by introducing a fine silver canula into their respective ducts.

In addition to the above-mentioned differences in the individual secretions of which the saliva of the dog is made up, Jacobowitsch further notices that, (a) the parotid saliva, exposed to the air, becomes rapidly covered with a film of crystals of carbonate of lime, which is not the case with either of the other secretions; (b) that at a temperature of  $100^{\circ}$ , the parotid saliva does not become turbid, whilst the other secretions, at least in a slight degree, become opaque; (c) that the parotid saliva, if boiled with nitric acid, and subsequently treated with ammonia, does not assume the yellow or orange tint which is developed when the secretions of the buccal mucous membrane and the submaxillary glands are similarly treated; and (d) that it is only in parotid saliva that carbonate of potash produces a slight precipitation of carbonate of lime.

Jacobowitsch has also analyzed the mixed saliva of the dog, in one instance with the exclusion of the parotid, and in another with that of the submaxillary secretion.

After this review of the chemical characters of the different secretions constituting the saliva, we have little to add regarding the composition of MIXED or ORDINARY SALIVA.

In the ordinary saliva of man, Berzelius<sup>1</sup> found 0.71% of *solid constituents*, Tiedemann and Gmelin<sup>2</sup> 1.14 to 1.19%, Wright<sup>3</sup> 1.19%, and L'Héritier<sup>4</sup> 1.35%; Jacobowitsch found only 0.484%; Frerichs in 18 analyses, 0.51 to 1.05%; and, from numerous determinations of filtered saliva, I have only found from 0.348 to 0.841%; so that the statements of the older observers are obviously too high. In the saliva of the dog Jacobowitsch found 1.037%, and in that of the horse Magendie and Rayer found about 1% of solid residue.

In 100 parts of the solid constituents of mixed human saliva, Tiedemann and Gmelin found 21.3% of fixed salts, while L'Héritier found only 6.8%, and Jacobowitsch, on the other hand, 37.5%; the last-named observer found that the mineral constituents predominated very much in the saliva of the dog, where they amounted to 65.5% of the solid residue; in the horse, according to Magendie, they amounted to 40%.

In relation to the individual *mineral constituents* of the saliva, we are as little able to arrive at any definite conclusion regarding those which exist preformed in it from the analyses of the ash of the salivary residue, as in the case of most of the other animal juices. We have, however, already remarked, that a great part of the *alkali* in the saliva is combined with ptyalin, from which it is separated by the weakest acids, as, for instance, carbonic acid. From the quantities of acids which are requisite for the saturation of alkaline saliva, Wright has concluded that in the normal state the alkali never amounts to 1% of the saliva. In the

<sup>1</sup> Föreläsningar i Diurkemien. 2 vol. Stockholm, 1808.

<sup>2</sup> Verdauung nach Versuchen, Bd. 1, S. 9 ff.

<sup>4</sup> Chimie pathol. p 290. Paris, 1842

<sup>3</sup> Op. cit.

ash of the salivary residuc the alkali is for the most part combined with *phosphoric acid*; thus Enderlin<sup>1</sup> found 28·122% of the tribasic, and Jacobowitsch 51·1% of the bibasic phosphate of soda in the ash.

We never find more than a trace, and often not that, of the *alkaline sulphates* in fresh saliva; and even in the ash they are not found in any considerable quantity; hence, as in the case of the phosphoric acid, the sulphuric acid must have been formed from other compounds during incineration.

In the ash of human saliva Enderlin found 21·35% of sulphate of soda; and in that of horses' saliva I found 1·604% of this salt.

In Wright's method of determining the sulphocyanide of potassium, which consists in dissolving in water the extract taken up by ether, and precipitating with basic acetate of lead, not only sulphocyanide of lead, but a far larger quantity of a compound of lead with a fatty acid, is thrown down; from this circumstance Wright's determinations are on an average ten times too high.

The *chlorides of potassium and sodium* especially preponderate over the other mineral constituents of the saliva.

Enderlin found 61·930% of alkaline chlorides in the ash of the saliva, and Jacobowitsch 46·2%; in the dog they amounted, according to the last-named observer, to 85·7%.

*Sulphocyanide of potassium* never occurs in the saliva except in very small quantity.

Jacobowitsch found 0·006% of this salt in his own saliva; and in analyzing my saliva I found it to vary between 0·0046 and 0·0089%; according to Wright it ranges in human saliva from 0·51 to 0·98%, which is obviously far too high.

Although we have already spoken of the existence of sulphocyanide of potassium in the saliva, yet the very dogmatical assertions of Strahl,<sup>2</sup> who denies that the presence of sulphocyanogen can be demonstrated, and applies to the experiments of his predecessors, such terms as "deficient, irregular, and ill-judged" (notwithstanding that Gmelin has exhibited pure sulphocyanogen in very large quantity from the saliva by distillation), compel us to refer to the admirable memoirs of Jacobowitsch and Tilanus,<sup>3</sup> who have demonstrated beyond all doubt the presence of sulphocyanogen by the simplest and most unquestionable chemical experiments; Frerichs<sup>4</sup> has also established the proof of the existence of sulphocyanide of potassium in the saliva. Moreover, it was formerly shown by Marchand,<sup>5</sup> and it has been more recently demonstrated by Wöhler and Frerichs,<sup>6</sup> by direct experiments, that sulphocyanide of potassium does not possess any poisonous properties.

Local stimulation of the salivary glands, the internal use of prussic acid and the salts of cyanogen, and especially of sulphur, increase, according to Wright, the quantity of sulphocyanogen in the saliva.

In mixed or ordinary saliva the *ptyalin* is mixed with mucus, so that

<sup>1</sup> Ann. d. Ch. und Pharm. Bd. 49, S. 817.

<sup>2</sup> Med. Zeitg. v. d. Ver. f. Preussen. 1847. Nr. 21 u. 22.

<sup>3</sup> De Saliva et Muco; dissert. inaug. Amstelod. 1849.

<sup>4</sup> Op. cit. p. 764.

<sup>5</sup> Lehrb. d. Phys. Ch. 1844, S. 410.

<sup>6</sup> Ann. d. Ch. u. Pharm Bd. 65, S. 844.

its properties do not appear to be altogether identical with those which we have described, and its accurate quantitative determination is impossible, independently of the considerable amount of salts contained in the aqueous extract. The aqueous extract, consisting chiefly of ptyalin, was found by Berzelius to amount to 40·8% of the solid residue of the saliva, while Gmelin fixed it at 20·0%, and Van Setten<sup>1</sup> at 15·62%.

The determinations of the organic matter soluble in water and in alcohol, are very uncertain; indeed, little or nothing is known regarding this substance.

The quantity of mucus in the mixed saliva must obviously be liable to very great variations, according to the conditions under which this fluid is collected.

The ether-extract ranged from 5·8 to 9·6% of the solid residue in the cases in which I determined it in several analyses of my own saliva.

In reference to the chemical *qualitative and quantitative analyses of the saliva*, it may be generally observed that the same principles and methods are applicable which have been described, in our remarks on the different animal substances; hence we need here only refer to a few points which require a special mode of treatment. In the first place the saliva must always be filtered, in order to effect the removal of the epithelial scales; unfortunately, however, the saliva is often so viscid and tenacious, that it undergoes decomposition before passing through the filter; indeed, it generally happens that the small quantity of filtered and perfectly clear fluid begins to become turbid, while the greater portion of the fluid still remains upon the filter. In such cases it is often advisable to dilute the saliva with three or four times its volume of boiling water; and after the mixture has been as thoroughly as possible cooled, and the mucous flocculi for the most part deposited, to filter and proceed with the analysis; but as in this case we are not able to separate the soluble from the insoluble portion, it is better not to attempt the whole analysis, since we should only obtain inaccurate results. We might certainly at once evaporate the viscid fluid, in order to extract the residue with alcohol, ether, and finally with water; but independently of the circumstance that the aqueous extract is also difficult of filtration, substances would be taken up by the alcohol and ether, which do not pertain intrinsically to the saliva, but to the epithelial cells, and to the fat and remains of food sometimes mixed with them.

It is obvious that if, before submitting the saliva to a chemical analysis, we duly examine its morphological elements with the microscope, we can ascertain whether the insoluble parts of the saliva consist merely of epithelial cells and mucous corpuscles, or whether they also contain fat, vibriones,\* or molecular granules of an organic nature. In saliva which has been for a long time exposed to the air, in morbid saliva, and especially when it exhibits an acid reaction, such granules are of very frequent occurrence. As substances for the most part in an actual state of change, they do not fall within the domain of an accurate chemical analysis. No one can confound mineral substances, as, for instance, crystals of carbonate of lime, with these molecular granules.

<sup>1</sup> Op. cit. p. 24.

If the saliva has been filtered, no interest attaches to any investigation of the residue left on the filter, at least in so far as the nature of the saliva is concerned, seeing that true saliva contains only soluble substances.

Wright finds his ptyalin in this residue; he cannot, however, possibly have treated this residue with sufficient water, since, in that case, it could not have contained so large a quantity of a matter soluble in water as his numbers indicate.

If carbonate of lime be mixed with this residue insoluble in water, it may easily be extracted by very dilute acetic acid, and its quantity subsequently determined.

In reference to the filtered solution, it is generally of interest to determine volumetrically the amount of acid which is saturated by a certain quantity of saliva, in order to form an opinion regarding the alkalinity of the saliva, or, in other words, regarding the quantity of the weakly combined alkali. In every case, however, the filtered saliva must be neutralized with acetic acid, and then heated; if this gives rise to a turbidity, the albuminous substance which is precipitated must be collected on a filter, and determined quantitatively. The residue left on the evaporation of the filtered saliva is then to be treated in the same manner as we treat the residue in the case of most of the other animal fluids.

It only remains for us to make a few remarks on the quantitative determination of the sulphocyanogen. The following are at present regarded as the two best methods of effecting this object. One method consists in dissolving the alcoholic extract of the saliva in water, and filtering the fluid, which is generally turbid from the presence of fat; the filtrate, after being somewhat concentrated by evaporation, is heated with phosphoric acid, and distilled; the distillate is saturated with baryta, and the filtered fluid evaporated; the residue is then boiled for a long time with fuming nitric acid or aqua regia, and the quantity of sulphocyanide of potassium is calculated from the amount of sulphate of baryta which is separated (Van Setten, Jacobowitsch, Tilanus). In following the other method, we first precipitate the aqueous solution of the alcoholic extract with nitrate of silver, and treat the well-washed deposit with water containing nitric acid, which leaves the chloride of silver undissolved; we then precipitate the silver from the acid solution with hydrochloric acid, add a little chloride of barium, and evaporate slowly, adding from time to time some nitric acid: in this way also we obtain sulphate of baryta, from which the sulphocyanogen must be calculated. Before the addition of the baryta, we should also be able to precipitate the sub-sulphocyanide of copper by the addition of the sulphates of protoxide of iron and oxide of copper; as, however, the precipitate never consists of pure sub-sulphocyanide of copper, we are compelled to determine the sulphur as sulphate of baryta.

The application of basic acetate of lead, according to Wright's method, for the precipitation of the sulphocyanogen, is inapplicable in this case; for the sulphocyanide of lead is somewhat soluble in water, and the greater part of it would probably be lost on washing.

*Abnormal constituents* occur in the saliva probably more frequently



than in many other animal secretions. It is very remarkable, that many mineral and organic substances which are thrown off by the urine either unchanged or little modified, are far more rapidly eliminated by the salivary glands—often, indeed, before they could be separated by the kidneys from the mass of the blood. We may very readily convince ourselves of this fact, by taking 5 grains of iodide of potassium in pills, when we shall find that it can be much sooner detected in the saliva than in the urine; in the latter fluid we may very often easily discover it after forty hours.

Moreover, when iodine is applied externally, as, for instance, in the form of ointment, it very rapidly passes into the saliva, where it may be recognized by nitric acid and a solution of starch, while it cannot be detected in the urine.

When iodine has been taken in the form of pills, and we have convinced ourselves, immediately after they have been swallowed, of the absence of this substance in the buccal mucus and in the saliva, we may very often detect it in the last-named fluid after a lapse of ten minutes, while it will not appear in the urine for a period varying from half-an-hour to two hours.

*Bromine* and *mercury*, and probably several other sialagogues, behave in this respect like *iodine*.

The reason why these substances so readily excite the flow of saliva, is probably solely dependent on the circumstance that they are separated from the blood by the salivary glands. It is possible that several organic sialagogues act simply in the same manner, namely, by some of their constituents, like the iodine, being readily separated by the salivary glands.

Wright and several other observers have been unable to detect any *mercury* in the saliva during mercurial salivation. I formerly had many opportunities of examining the saliva in cases in which salivation ensued during the treatment of syphilis by inunction practised by Rust and Louvrier, and I constantly found mercury in this fluid, both by dry distillation of the residue of the saliva, and by the simple application of an extremely small pair of plates of copper and zinc to the slightly acidified saliva. There are many reasons why, even when much mercury had been taken into the organism, none was found in the saliva; in the first place, it has very often been only buccal mucus which has been examined, for we may readily convince ourselves by the microscope, and more accurately by chemical analysis, that at the commencement of salivation scarcely any saliva is found in the sputa; the salivary glands are as yet not affected; the expectoration consists almost entirely of whole patches of epithelium and of mucus from the tonsils; and in products of this nature I have never been able to detect any mercury, even after its free administration; and, secondly, it has been forgotten by some experimentalists, that mercury volatilizes very readily with the vapor of water, so that, by evaporating too rapidly, and without sufficient care, they have allowed the little mercury that was present to escape.

Wright has injected alkaline carbonates into the veins of animals, and has found a consequent augmentation of the alkali in the saliva; when, on the other hand, he injected acetic acid or extremely diluted sulphuric

acid into the vessels of healthy animals, he never found that an acid reaction of the saliva was induced.

It is singular that dogs into whose jugular veins Wright injected four ounces of pyroligneous acid, and half a drachm of sulphuric acid (although the acids were diluted with four and six ounces of water), bore this outrage so well, that after a short time they quite recovered,<sup>1</sup> and Wright found that their saliva had returned to its alkaline reaction. In cases in which I have performed similar experiments on animals, although for a different object, death was the invariable result—an event which may be very easily explained, since stasis must be very rapidly induced in the pulmonary capillaries, in consequence of the coagulation or gelatinizing of the blood.

We have already referred to the incidental occurrence of *sugar* (p. 257) and of lactic acid (p. 94) in the saliva. It is extremely difficult to decide whether actual albumen coagulable by heat is present in a specimen of saliva.

Wright assumes that there are two different kinds of albuminous saliva, and as we have no experience on the point, we cannot contradict his assertion; but as he also asserts that albumen occurs in normal saliva, which is certainly not the case, at least in any appreciable quantity, and as further, the recognition of small quantities of albumen is difficult and often impossible, for the reasons before given, we are justified in doubting whether albuminous saliva is of frequent occurrence.

*Biliary matters*, and especially cholesterin, sometimes pass, according to Wright, into the saliva. (See p. 120.)

Wright has described a large number of varieties of saliva, classifying them according to the heterogeneous constituents which they contain; thus he distinguishes ammoniacal saliva, saliva containing hydrochloric acid, saline saliva, puriform saliva, bloody saliva, fetid saliva, &c. In declining to adopt Wright's results in our "Physiological Chemistry," we by no means wish to detract from the meritorious portion of his careful labors; but we do not think that the substrata on which such investigations are founded are of a nature to rank high in exact sciences such as chemistry and physiology. Descriptions of the subdivisions of morbid saliva, as for instance, of bilious, bloody, puriform, fetid, acrid, frothy, and gelatinous saliva, may have a certain importance in relation to semeiotics, but cannot serve as substrata for physiological inquiry. The illogical character of such a classification is obvious; the chemical investigations often do not justify the conclusions which Wright has drawn from them; for sugar, bile, lactic acid, &c., are never recognized by him with such certainty as chemists of the present day require; moreover, recent physiology might require further particulars regarding acrid, puriform, and bloody saliva, while our present pathology pays less attention to ontological ideas of disease than to investigations founded on actual physical diagnosis and on morbid anatomy. We repeat that we by no means wish to ignore the many interesting facts with which science has been enriched by Wright's rich experience and indefatigable labor.

<sup>1</sup> [This is hardly correct. The dog into whose vein the pyroligneous acid was injected, died in about a week. See the "Lancet," 1842-3, vol. ii. p. 189.—G. E. D.]

Although *acid saliva* has been observed in a large number of instances, our knowledge of it is still very defective, for notwithstanding the positive assertions of Wright, there is as yet no proof that lactic acid is the cause of this acid reaction. Moreover, Prout<sup>1</sup> has adduced no decisive proof of the presence of this acid.

We have already shown, that the acid reaction of the animal fluids may depend on the presence of other acids (as for instance, several of the butyric acid group), or even of acid phosphate of soda: hence it is invariably necessary to determine the nature of the free acid, whenever it is present in the saliva, in different diseases, before we venture to decide regarding the course of the chemical process accompanying the disease; it is, however, the chief aim of all chemical investigations of animal objects, to draw from them a conclusion regarding the nature of the chemical process in the healthy or diseased state. For semeiotics the simple statement suffices that in this or that condition the saliva exhibits an acid reaction. We shall now briefly mention those pathological states in which, as yet, the saliva has been found to present an acid reaction.

The saliva is *acid*, according to Donn  ,<sup>2</sup> in inflammatory affections of the *prim   vi  *, in pleuritis, encephalitis, acute rheumatism, intermittent fever, and uterine affections, and, according to L'H  ritier, also in cancer of the stomach. Wright assumes that there are four varieties of acid saliva, namely, (a) that which occurs in idiopathic affections of the salivary glands; (b) that which presents itself when there is a predominance of acid in the organism generally, from constitutional or other causes, amongst which he mentions scrofula, phthisis, rachitis, amenorrh  a, inflammatory rheumatism, &c.; (c) the form occurring in subacute inflammation of the mucous membrane of the stomach and intestines; and (d) the form presenting itself in dyspepsia (a somewhat vague mode of expression). In affections of the nervous system, the saliva is on the other hand never acid, but often very strongly alkaline. In catarrhal affections of the gastric and intestinal mucous membranes, and in the round perforating form of ulceration of the stomach, I have very often, but not invariably, found the saliva acid; in cancer of the stomach and in diabetes, I have, however, always found it acid. In inflammatory affections of the thoracic organs, in acute rheumatism, typhus, &c., I<sup>3</sup> have very often found the saliva alkaline or perfectly neutral. According to Donn   and Frerichs<sup>4</sup> the acid reaction always depends on the buccal mucous membrane, which, when in a state of abnormal irritation, invariably yields an acid secretion.

Amongst the difficulties which usually present themselves in the investigation of morbid saliva, we may notice that we can rarely obtain a quantity sufficient for analysis, seeing that it is a fluid which contains only a very small amount of solid constituents. Hence it might be expected that it would, at all events, have been more accurately examined in persons with pytalism, in which it is secreted in large quantities, but such is not the case. Wright has indeed given an exposition of those

<sup>1</sup> On the Diseases of the Stomach, 4th ed. p. 451.

<sup>2</sup> Histoire physiol. et pathol. de la Salive. Paris, 1836.

<sup>3</sup> Schmidt's Jahrb. Bd. 36, S. 185.

<sup>4</sup> Op. cit. p. 761.

cases in which a symptomatic, a critical, or an artificially induced ptyalism occurs; but we do not the less miss analyses susceptible of chemical and physiological application. The secretion in cases of *mercurial salivation* has as yet been the most carefully studied. The results obtained by Wright, L'Héritier, Simon, and myself, perfectly coincide in the following points. At the commencement of mercurial salivation the buccal mucous membrane and the tonsils are more affected than the salivary glands, and hence the expectoration is more viscid, of a higher specific gravity, and richer in solid constituents, especially epithelium and mucus-corpuscles, than normal saliva; it is very turbid, from the presence of more or less distinct flocculi; has an alkaline reaction; contains little of the peculiar principle, ptyalin, often much fat, and rarely any appreciable quantity of sulphocyanide of potassium. At a later period, when the salivary glands become the seat of pain and swelling, a less turbid saliva is secreted, which often contains a much smaller amount of solid constituents than normal saliva; it is still alkaline, and sulphocyanide of potassium is more often absent than present; it is also rich in fat, and often in mucus-corpuscles. We have already noticed the presence of mercury in this variety of saliva.

In conclusion, we must mention *salivary calculi* as morbid products of this secretion: they have been very often analyzed, and have been found to contain more carbonate of lime than any other kind of concretion. As we have already shown that the saliva even of carnivorous animals holds in solution no inconsiderable quantity of lime in combination with organic matter, and that the lime is very readily separated from this compound as a carbonate, we have no difficulty in comprehending the mode of formation and the constitution of these concretions.

*The quantity of saliva excreted in a given time* is a point regarding which there is a tolerable coincidence amongst the statements of authors, although we cannot regard it as definitively established; for most writers have based their calculations on the data of Mitscherlich, which refer merely to the parotid secretion of a man laboring under disease.<sup>1</sup> The patient in the case referred to, after collecting the saliva in the mouth for 15 minutes, ejected from it 6·27 grammes, while, during the same period, 0·92 of a gramme were discharged by the fistula; moreover Mitscherlich never determined the quantity of the parotid secretion, except under definite relations and in given times. Now if we assume that the above ratio of the parotid secretion to the secretion of the other salivary organs is constant, which, however, is more than doubtful, we may calculate the quantity of saliva which will be secreted in a definite time, or under definite physiological relations. Under ordinary circumstances (that is to say, on the common hospital diet) Mitscherlich found that the quantity of the parotid saliva in 24 hours varied from 46·3 to 74·8 grammes. Assuming the above ratio of the parotid secretion to that of the other sources of the saliva to be constant, the whole amount of saliva from the six salivary glands and the buccal mucous membrane, would amount on an average to 473 grammes in 24 hours. Burdach<sup>2</sup> calculates, from Mitscherlich's data, that the saliva secreted by an adult in 24 hours

<sup>1</sup> [Lehmann should have mentioned that in this case the patient had a parotid fistula.  
—G. E. D.]

<sup>2</sup> Physiol. Bd. 1, S. 277 ff.

amounts to 10 (German) ounces (= 255 grammes). Valentin<sup>1</sup> assumes the quantity at from 216·4 to 316·3 grammes; Donnè<sup>2</sup> fixes the quantity at 390 grammes, and Thomson<sup>3</sup> at only [3216 grains or] 210 grammes.

Jacobowitsch has determined in dogs the quantities of saliva which he could collect from each set of salivary glands in an hour; from the two parotids he obtained 49·2 grammes, from the submaxillary glands 38·83 grammes, and from the orbital and sublingual glands, and the buccal mucous membrane, 24·84 grammes. We can draw no conclusions from these data, regarding the quantity of saliva secreted in a normal state in a definite time; for independently of the circumstance that nothing is stated regarding the size or the weight of the dog, we know from his numerical statements, that Jacobowitsch employed dogs of various sizes in his experiments, so that the fluid was collected under such peculiar conditions, that a comparison of it with the quantity of the secretion in the normal state is impossible. Jacobowitsch, however, arrived at the interesting result, that to whatever extent the quantities of the saliva secreted by the different organs may vary, the solid constituents—both the organic and the inorganic substances—amount to very nearly the same from all three of the sources; thus, in the quantities above given of parotid, submaxillary, and sublingual saliva, the solid constituents amount to very nearly the same, that is to say, to about 0·232 of a gramme, of which 0·080 is organic and 0·152 inorganic matter.

All determinations of the quantity of saliva secreted during a more prolonged interval (as, for instance, 24 hours), must at most be regarded as merely approximative, since the activity of the salivary organs is dependent on very different influences and conditions. The most common cause exciting a copious discharge of saliva is the mastication of food; it depends, however, very much on the nature of the food, whether much or little saliva is effused into the buccal cavity; dry and hard food inducing a more copious flow of saliva than food which is moist and soft; indeed, the mere motion of the lower jaw excites the act of secretion, and hence, speaking or singing is always accompanied by the secretion of saliva. That chemical irritants, such as are present in acid and aromatic articles of food, and mechanical irritation, such as tickling the palate, often produce an immediate secretion, is as well known as that certain psychical influences always produce a similar effect. It is especially important to observe that after the use of food, the secretion continues for a long time; a phenomenon which appears not to be so referable to the irritation transmitted to the salivary glands from the buccal cavity, as to the communication of nervous action from the stomach during the process of digestion; for, on introducing food into the stomach, either through a gastric fistula, or by means of an elastic tube in the œsophagus, we observe that the secretion of gastric juice is accompanied by a copious effusion of saliva.

In order to determine the quantity of saliva required for different kinds of food, experiments have been instituted by Magendie and Rayer,<sup>4</sup> by Lassaigne,<sup>5</sup> and by Bernard,<sup>6</sup> on horses. The œsophagus

<sup>1</sup> *Physiol. d. Menschen*. 1844, Bd. 1, S. 626.

<sup>2</sup> *L'Institut*, No. 158, p. 59.

<sup>3</sup> *Animal Chemistry*. Lond. 1848, p. 571.

<sup>4</sup> *Compt. rend.* T. 21, p. 902.

<sup>5</sup> *Journ. de Chim. Méd.* 1845, p. 472.

<sup>6</sup> *Arch. gén. de Méd.* 4 Sér. T. 13, p. 22.

was exposed and opened, and the food which the animals had swallowed was intercepted and removed. From these experiments it followed, that straw and hay, as they pass down the oesophagus, are mixed with four or five times their weight of saliva, whilst seeds abounding in starch, as, for instance, oats, are mixed with an equal quantity, or perhaps one and a half times as much of saliva, and fresh green fodder with only half its weight; and that food mixed with water seems to take up scarcely any saliva. Hence it appears as if, when food is taken, the secretion of saliva is only dependent on its nature, and as if, when fluid or moist food is taken, the glands are not excited to activity. We must, however, assume with Frerichs, that even in a state of perfect repose, the secretion of saliva is not totally suspended; for although Mitscherlich found scarcely a trace of saliva escaping from the fistulous openings in the patients on whom he experimented, when they had fasted for some time and were in a state of perfect repose, and although we observe scarcely any secretion in horses in whom a fistulous opening has been established, when they have not been supplied with food for some time, we cannot suppose that the process of secretion is absolutely suspended in these any more than in other glands. Moreover we can form no opinion regarding the normal secretion in a state of relative repose, from the facts that during sleep, when the head is inclined forwards, and in paralysis of the facial muscles, saliva is secreted in no sparing quantity, since in both those cases the abundance of the secretion is dependent on peculiar conditions.

Colin<sup>1</sup> appears to have made very extensive observations on the secretion of saliva in the solidungula. Amongst other results of his investigations he mentions that the secretion of the two parotids alternates, the parotid of the side on which mastication is going on, secreting at least one-third more than that of the other side; and that when the masticatory process is transferred to the other side, the activity of the first gland very rapidly diminishes, and that of the second as rapidly increases. He did not observe the alternating action in the secretion of the submaxillary glands, which is apparently uniform on both sides. When the animal consumes dry food, there are secreted from 5000 to 6000 grammes of saliva from all the glands in the course of one hour; about 1-3d or 1-4th more when the animal consumes oats; and 1-5th or 1-4th less when living on succulent roots. The parotids alone yield more than 2-3ds of the whole sum, the submaxillaries only 1-20th, and the sublinguals and mucous follicles the remainder. The secretions of the parotid and submaxillary glands occur almost solely during mastication, and for a short time subsequently; the thick and tough secretion of the other glands of the buccal cavity, which remain moist during abstinence, amount to only 1-37th of the whole. The sight of food excites no perceptible augmentation of the salivary secretion even in fasting animals.

Some very interesting experiments on the influence of the period of secretion on the chemical constitution of the saliva, have been made by Becher and Ludwig.<sup>2</sup> They found that the solid residue of the saliva diminishes in proportion to the amount which the gland has already

<sup>1</sup> Compt. rend. T. 34, pp. 827-830.

<sup>2</sup> Zeitschr. f. rat. Med. N. F. Bd. 1, S. 480-483.

yielded; the organic constituents sinking far more rapidly than the inorganic. Fluctuations in the quantity of water in the blood did not disturb this law, as was proved by the examination of saliva collected after one or more venesections; nor was it affected by the injection of chloride of sodium into the blood, although the quantity of salts in the saliva was somewhat augmented thereby.

Physiologists have ever held the most varied opinions regarding the *physiological value* of the saliva. We must, however, regard the saliva as essentially fulfilling a threefold object of a mechanical, a chemical, and a dynamical nature.

The *mechanical* object is so manifest, that no one has ever seriously doubted it; for it is obvious to our senses, and requires no demonstration to convince us that the moistening of dry food in mastication serves, on the one hand, the better to adapt it for deglutition, and, on the other hand, to separate the particles, and thus allow them the more freely to be acted on by the other digestive fluids. Formerly, however, the whole value of the saliva was limited to this function; and Bernard recently believed that he had proved this view to be correct by the experiments to which we have alluded. He maintained that the parotid saliva, by its thin fluid property, serves to moisten the food, while the tough and viscid secretion of the submaxillary glands affords a mucous investment to the masticated food, lubricates it, and thus adapts it for deglutition.

We have already seen that the secretion of the submaxillary glands is distinguished by its viscosity and toughness from the parotid secretion, and even that infusions of these glands differ in the same manner. In reference to this circumstance Bernard notices the fact in comparative anatomy, that those animals which swallow their food without masticating it, as, for instance, serpents, birds, and reptiles, possess no parotid glands, or at most only rudimentary organs, while their submaxillary glands for the elaboration of mucus are for the most part very well developed.

The *chemical function* of the saliva is a subject on which different observers have held very different views. Leuchs was the first who was led by experimental inquiry to the discovery that starch is gradually converted into sugar by the action of the saliva. Subsequent inquirers who have repeated the experiments in some instances confirm, and in others deny, the accuracy of his views. Wright, who bases his view on a large number of experiments, speaks very decisively in favor of the chemical action of saliva on amylaceous food; and indeed, Mialhe<sup>1</sup> believed that he had actually discovered the substance in which this metamorphic power solely or for the most part resides, and gave it the name of *salivary diastase*. Several observers, having failed in attempting confirm the views of Wright and Mialhe, have directed their attention to the individual secretions of the different glands. Magendie was the first who discovered that neither the parotid nor the submaxillary secretion exerts any action on starch, but that the common or mixed saliva of the horse converts both crude and boiled starch into sugar at the temperature of the animal body; Bernard attributed this unquestionable property of the mixed saliva (whether obtained from man, the dog, or

<sup>1</sup> Compt. rend. T. 20, pp. 247, 367, 954, et 1485.

the horse), solely to the buccal secretion, while Jacobowitsch has adduced convincing evidence that this secretion alone does not possess the above property which exists only in the mixture (whether artificial or natural) of the secretions of the buccal mucous membrane and the salivary glands. It can therefore no longer be doubted that the saliva, in the normal condition in which it is mixed with the food, possesses the property of converting starch into sugar. The more recent carefully conducted experiments of Bidder and Schmidt<sup>1</sup> have, however, shown that the parotid secretion does not contribute to the action of the mixed saliva. Parotid saliva and buccal mucus do not metamorphose starch, although this effect is rapidly produced by the secretion of the submaxillary glands and the buccal mucus. These inquirers arrived at the same result, namely, that the starch-ferment is only developed by the union of the buccal mucus with the submaxillary saliva, by tying the ducts of the different salivary (the parotid and the submaxillary) glands in dogs. We by no means, however, agree with Ross<sup>2</sup> in regarding the buccal cavity as the stomach for vegetable food. Even under the most favorable circumstances we cannot detect a trace of sugar in a mixture of saliva and boiled starch, in less than from 5 to 10 minutes; and hence, if the conversion of amylaceous matter into sugar be the physiological function of the saliva, its action must not be confined to the short time in which the food remains in the mouth, but must also be continued in the stomach and intestines. Now we may readily convince ourselves that this is really the case, by observing what occurs in an animal in whom a gastric fistula has been established; for while pure gastric juice exerts no action on starch, sugar may be detected by the ordinary means in the stomach of the animal in 10 or 15 minutes after it has swallowed balls of starch, or after they have been introduced through the fistula. Hence it cannot be doubted that the saliva, after it has been mixed with the other animal secretions, continues to exert its action on the *amylacea* in the digestive canal.

Notwithstanding the evidence which has been adduced to prove that the saliva exerts this action on starch, there are other facts which seem to show that we must not assign to it an extreme importance in the digestive process in general. For, even if we do not admit the conclusiveness of Budge's<sup>3</sup> experiment, in which he extirpated all the salivary glands of a rabbit, and afterwards observed no disturbance of the digestive system, and no imperfection in the nutrition, yet on the following grounds we must regard this action of the saliva as a somewhat limited one: the quantity of the saliva which is secreted is altogether independent of the quantity of starch contained in the food, and is extremely small when the latter is taken in a fluid form; when food which has been thoroughly moistened is swallowed, there is only a very slight secretion of saliva; liquid and moistened foods remain in the stomach so short a time, that a perfect conversion of the starch into sugar in this organ is impossible; nature has, however, provided a secretion which is poured into the duodenum—the pancreatic juice, which possesses the power of effecting this conversion of starch into sugar in a far higher degree than

<sup>1</sup> Die Verdauungssäfte und der Stoffwechsel. S. 21.

<sup>2</sup> The Lancet, January 18, 1844.

<sup>3</sup> Rhein. Blätt. Bd. 4, S. 15.



the saliva; animals (fishes, for instance) which swallow amylaceous food without masticating it, possess for the most part such rudimentary salivary glands, that the secretion from them can hardly be taken into consideration. But even the pancreatic juice is not generally sufficient to effect the perfect metamorphosis of the starch; the conversion proceeds so slowly that we can almost always detect a considerable quantity of starch in the excrements not only of carnivorous but also of herbivorous animals, after the ingestion of amylaceous food. Hence it appears very much to depend on the subjective opinions of different writers, whether a greater or lesser importance in this point of view be attributed to the saliva; in no case, however, should the function of the saliva as a saccharifying agent be overrated.

This is a subject on which special observations, experiments, and criticism, are peculiarly needed; and it is by the neglect of this mode of inquiry that some of the best observers have been erroneously led to adopt the most extreme views. The difficulty of forming a decided judgment without a special investigation will be best seen from the following historical sketch of the facts which have been accumulated in reference to this subject. Wright,<sup>1</sup> whose views are based on a large number of experiments, is one of the strongest supporters of the digestive powers of the saliva; and I<sup>2</sup> formerly entirely coincided in this view; but all experiments and results bearing on this point must only be adopted with the greatest caution, for there is no analytical inquiry in which, under apparently precisely similar relations, the same experiment so often yields different results, and in which quantitative determinations so invariably present a want of uniformity. Thus, the quantities of starch converted into sugar in contemporaneous experiments with the same saliva, and at perfectly equal temperatures, are often extremely different. Even when there is only a very small quantity of starch in relation to the saliva, we almost always find that the whole of it has not undergone conversion into sugar, as was observed by Jacobowitsch; it is only after a very considerable time, seldom before from 16 to 24 hours have elapsed, that we find the whole of the starch changed; and then the starch is not merely converted into sugar, but this latter substance has already undergone further changes, and has given rise to the formation of lactic acid—a change which often commences while very large quantities of starch still remain unchanged. We must further bear in mind that many other animal substances under certain conditions possess a similar power of converting starch into sugar. Liebig long ago observed that gelatin and albuminous and gelatigenous tissues, when they had been lying in a state of moisture, and exposed for some time to the atmosphere, possessed the property of effecting this change. Magendie<sup>3</sup> subsequently convinced himself that infusions of cerebral tissue, of heart, liver, lungs, and spleen, possessed to a certain degree the power of converting starch into dextrin and sugar; he likewise found that the serum of blood at 40° possessed this property, and that boiled starch was converted into sugar even in the circulating blood of the living animal. Hence Bernard<sup>4</sup> merely repeated the experiments of

<sup>1</sup> Op. cit.

<sup>2</sup> Schmidt's Jahrb. Bd. 37, S. 121–123, Bd. 39, S. 155 ff.

<sup>3</sup> Compt. rend. T. 23, pp. 189–192.

<sup>4</sup> Arch. gén. de Méd. 4 Sér. T. 13, p. 10.

Liebig and Magendie, when he exposed well-prepared and cleaned buccal mucous membrane to the air, and subsequently observed its action on starch. These facts cannot, however, be placed in comparison with the action of the mixed saliva, which does not require any long exposure to the atmosphere in order to acquire this property, and is only exceeded in this power by the diastase of germinating seeds and by the pancreatic juice.

Another point which must appear doubtful to those who have not experimented for themselves, is the question whether acid saliva has the same saccharifying force as the alkaline secretion—a view most positively denied by Sebastian, Wright, and Bernard; and as confidently asserted by Jacobowitsch and Frerichs. In my former experiments I failed, like the first-named observers, in effecting the conversion of starch by saliva, and notwithstanding the most careful inquiry, I have been unable to detect the causes of that failure; but in my more recent experiments, when I have allowed saliva treated with acetic, sulphuric, hydrochloric, or nitric acid, to act on either crude or boiled starch, I have always observed a tolerably rapid formation of sugar, and have convinced myself that acids can no more impede the digestive power of the saliva than alkalies can promote it. It is therefore certain that mixed saliva, whether it be alkaline or acid, acts on starch with equal energy at equal temperatures. Trommer's sugar-test cannot be directly applied in this investigation, for Frerichs has shown by an interesting experiment that suboxide of copper is immediately separated when saliva and starch are boiled with potash and sulphate of copper; we must, therefore, remove any starch or dextrin that may remain in the filtered fluid, by treating it with alcohol, before applying Trommer's test, or we must adopt some other means of demonstrating the presence of sugar in the mixture.

The albuminous substance occurring in the saliva, to which Mialhe has given the name of *Diastase salivaire*, is undoubtedly similar in many respects to diastase; but, on a rigid examination, the two substances are found to be altogether different. Mialhe obtains this salivary diastase by precipitating human saliva with absolute alcohol. On referring to the composition of saliva, it is easy to perceive that the substances which will be precipitated by alcohol are chiefly ptyalin and mucus with a quantity of salts, and it is in the mixture of these substances that Mialhe thinks that he has found the active principle of the saliva. In experiments with this mixture, I have altogether failed in obtaining evidence of the extraordinary powers which were attributed to it by Mialhe (who maintains that 1 part can rapidly effect the metamorphosis of 8000 parts of starch at a temperature of 37°); and although I formerly believed that I had obtained a somewhat similar result, I have since convinced myself that the metamorphosing force is neither concentrated in the admixture of substances indicated by Mialhe or by myself, nor yet in any other part of the extractive matters of the saliva. In the mean time, it would be unscientific to neglect all inquiry regarding this peculiarity of the saliva, or rather of one of its constituents, and to rest satisfied with the fiction that all excitors of fermentation are substances undergoing changes, and that such substances are

incapable of being submitted to chemical exhibition and investigation. All fictions that close the door on inquiry are to be rejected, unless they admit of a logical justification. If Schwann, Wasmann, and others, had remained satisfied with the conviction that the cause of the digestive power of the gastric juice did not admit of being investigated, we should not have advanced very far in the knowledge of the process of digestion. We can hardly condemn an inquiry into the hypothetical diastase of the saliva as absurd; for the saliva does not lose this property either by the action of heat or alcohol, and pepsin similarly retains its power even after having previously been combined with salts of lead. This much, however, is certain, that the ptyalin obtained by Berzelius, Gmelin, and Wright, was found in all cases to be deficient in the power of converting starch into sugar.

After it had been demonstrated, as already observed, first by Magendie, and subsequently by Bernard, that the secretions of some of the salivary glands did not exert any metamorphic action on starch, Jacobowitsch, under the direction of Bidder and Schmidt, prosecuted some admirable experiments on this subject, which we do not think it will be irrelevant to notice at some length in the present place. He convinced himself, by hindering the flow of the secretions of the parotid and submaxillary glands from entering into the mouth of a dog, that the mere secretion of the mucous membrane of the mouth (contrary to Bernard's assertion) was unable to convert starch into sugar. But when he tied the ducts of only a single pair of glands (excluding only the secretion from the parotid or that from the submaxillary glands), and suffered the dog to recover after the operation, and then, according to Bernard's method, as already described, digested starch with the saliva exuding from the open and depressed mouth of the dog, some of the starch was converted into sugar in the course of five minutes. The starch was also quickly metamorphosed when brought in contact with an artificial admixture of the above-mentioned glandular secretions and buccal or even nasal mucus. (The nasal mucus alone did not possess this property.) A mixture of the secretions of the parotid and submaxillary glands, without any secretion from the mucous membrane, was entirely deficient in this property.

To prove that the action of the saliva on starch is continued in the stomach, the same author instituted the following experiments: in one case he fed a dog in whom a gastric fistula had been established, upon boiled starch after a twelve hours' fast; repeated experiments showed that the contents of the stomach which were discharged from the fistula contained sugar. In another case Jacobowitsch introduced boiled starch through a fistulous opening into the stomach of a dog, in whom the excretory ducts of the salivary glands had been tied; but here he could not discover any trace of sugar in the contents of the stomach after a prolonged period.

Bidder and Schmidt, under whose superintendence the experiments of Jacobowitsch were instituted, have convinced themselves by later experiments, that the saliva loses its action on starch in the stomach of the living animal. They introduced boiled starch under the most varied conditions, into the stomachs of dogs through gastric fistulae, and found

that after two hours' retention in the stomach, at most only mere traces of sugar could be detected, while externally to the organism this metamorphosis always occurred, even when an excess of gastric juice was added. This perfect suspension of the action of the saliva on starch within the stomach cannot be sufficiently explained either by the comparatively short retention of the starch in the stomach, or by the assumption that the salivary diastase is digested by the gastric juice. For on the one hand the amylacea generally remain a sufficiently long time in the stomach to undergo metamorphosis, and, on the other hand, the gastric juice would also digest the salivary diastase externally to the organism, which is not the case. These results of Bidder and Schmidt may be to a certain degree explained by the assumption, that in these experiments (in which starch was introduced through a fistula, or in the form of very moist starch-paste, through the mouth) only little saliva flowed into the stomach, where it became too much diluted by the gastric juice.

We are, consequently, led by the earlier observations of Bernard, as well as by the more recent investigations of Bidder and Schmidt, to the conclusion, that notwithstanding its energetic action on starch, and notwithstanding its abundant supply, the saliva takes no very important part in the digestion of the amylacea. Hence its principal use in the animal body must be of a mechanical nature. Besides the uses of this nature, mentioned in the text, Bidder and Schmidt believe that one of the main objects of the salivary secretion is its co-operation in the perpetual interchange of the watery fluids within the living organism.

Wright attaches a very high degree of importance to the alkalinity of the saliva both during insalivation and during gastric digestion, and ascribes to it a second chemical function, viz., that of saturating the excessive quantity of acid introduced into the stomach or formed within it. It is certainly an undeniable fact that alkaline saliva is secreted after the ingestion of acid food; but this also occurs when highly seasoned food, spirituous liquors, and other stimulants have been taken, which cannot have been saturated or combined, like the acids, with the alkali of the saliva. It is extremely doubtful whether the object of this secretion is to saturate the free acid, since our experiments have in general rather tended to show that an excess of acid in the stomach is less injurious to the digestion of nitrogenous substances than any deficiency in its quantity. For the present, therefore, we must rest satisfied with admitting that as the saliva constantly becomes alkaline during or after eating, even in those cases in which it was acid before the ingestion of food, and as moreover its alkalinity increases after taking indigestible or acrid substances, the alkali probably contributes to promote the function of the saliva, although we must leave it to future inquirers to determine the manner in which this object is effected.

Wright supports his assertion by an appeal to his own experience; he found that the effect of spitting, after having partaken of a full meal, was always to induce an abundance of acidity with much pain in the stomach, and a corresponding alkalinity in the saliva.

The saliva exerts no metamorphic action on any of the carbohydrates excepting starch: cane-sugar, gum, vegetable mucus (bassorin), and

cellulose, remain unchanged in the saliva; it is only in certain species of sugar, and after long-continued digestion at a high temperature, that we observe the formation of lactic and subsequently of butyric acid.

The saliva exerts no action whatever on albuminous and gelatinous food; its utmost effect being to relax their tissues like pure water, and thus to render them more accessible to the action of the gastric juice.

Wright thought he had convinced himself from numerous experiments that flesh is softened and rendered tender in its texture much more rapidly when digested with saliva, than when it is subjected to the action of water; he further concluded from these and similar experiments, that the saliva contributes essentially to the digestion of animal substances; but Jacobowitsch and Frerichs have recently shown by their more accurate and well-tested experiments, that this view is utterly erroneous.

Bernard and Barreswil<sup>1</sup> believed that they were justified from some of their experiments in laying down the following proposition: "Le suc gastrique, le fluide pancréatique, et la salive, renferment un même principe organique, actif dans la digestion: mais c'est seulement la nature de la réaction chimique, qui fait différer le rôle physiologique de chacun de ces liquides, et qui détermine leur aptitude digestive pour tel ou tel principe alimentaire."

If this view had not been fully controverted by the admirable experiments of Jacobowitsch and Frerichs, its untenability would have been manifest to every one on a mere repetition of Bernard and Barreswil's experiments.

Liebig has suggested that the saliva may be designed from its tendency to frothing, to convey atmospheric air into the stomach and intestinal canal. Wright and others subsequently to him have shown that starch is metamorphosed by saliva obtained by expectoration (which has consequently been sufficiently exposed to the action of the air), without further access of oxygen; and Valentin<sup>2</sup> has very correctly stated that oxygen is not necessary for the digestion of animal substances by the gastric juice—facts which have been advanced in refutation of Liebig's view; but it should be borne in mind that these experiments were not conducted with such accuracy as to exclude all access of oxygen, and that they cannot therefore be advanced as sufficient evidence against the accuracy of Liebig's view: there are, moreover, as we know, certain processes, as for instance the vinous fermentation, in which it requires the greatest exactitude of observation to demonstrate the necessity of a slight access of oxygen. Then again, the fact that only mixed saliva, that is to say, saliva which has been in contact with atmospheric air, is capable of metamorphosing starch, speaks rather in favor of Liebig's view than against it. Even if the oxygen, which undoubtedly passes into the *primæ viæ* with the saliva, exerts no effect upon the process of digestion in the stomach, the use of this gas in the intestinal canal may readily be understood, although it cannot be specially demonstrated. We know that gases are present in the intestinal canal, and that these gases are rich in carbonic acid and often also in hydrogen compounds. The formation of the latter, whose passage into the blood would be followed by very in-

<sup>1</sup>Compt. rend. T. 21, p. 88.

<sup>2</sup>Lehrb. d. Physiol. des Menschen. Bd. 1, S. 286.

jurious results, must necessarily be greatly limited by the presence of free oxygen. According to the laws of the diffusion of gases, the presence of oxygen in the intestines must diminish the withdrawal of oxygen from the blood and the supply of carbonic acid and hydrogen to that fluid.

Wright considers that one of the most prominent functions of the saliva is its supposed property of serving as a necessary, *stimulant to the stomach*, and thus promoting the digestive process. We have already frequently expressed our dissent from the *dynamical* explanation of physiological phenomena; according to our view, even the nerves cannot act independently of chemical changes, and if we are to admit the control of dynamical forces on the nervous system, we must first establish the existence of definite chemical relations in proof of such an action. It appears to us altogether inconsistent to attach any importance in physiological chemistry to the obscure idea of an *irritant*. When I introduced fresh saliva, through a fistulous opening, into the stomach of a dog, I observed that the same amount of gastric juice was secreted as when other mucous fluids were conveyed into the stomach. There is no indication of any special irritant in experiments of this kind; and the stimulating action of the saliva can hardly be required for the process of gastric digestion, since solid substances, and more especially nitrogenous food, induce a far more abundant secretion of gastric juice than pure saliva.

Wright introduced from 3 to 10 ounces of saliva, through an elastic-gum catheter, into the stomachs of dogs that had been kept fasting, and observed, after the lapse of ten minutes, contraction of the abdominal muscles, uneasiness, eructations, and vomiting. I did not perceive any of these phenomena when I introduced fresh human saliva into the stomach of a dog, through a fistulous opening, but I certainly did not employ more than two ounces at most; six ounces of the parotid saliva of a horse were, however, equally well retained by the dog. Nor can any conclusions be deduced from vomiting in dogs, since they vomit on the slightest provocation, and frequently devour what they have thrown up without experiencing any bad effect from it. The quantity of saliva which was used by Wright, and which could not have been very speedily collected, leads us to suspect that his "normal saliva" was already undergoing decomposition, and consequently gave rise to these abnormal phenomena.

Wright also distinguishes several passive functions of the saliva; (a) it assists the sense of taste; (b) it favors the expression of the voice; (c) it clears the mucous membrane of the mouth, and moderates thirst.

We must not omit to mention, at the close of these remarks on the saliva, that Wright believes he has confirmed, by his experiments of injecting the saliva of animals into the blood, the ancient opinion, which, however, is still maintained by Eberle<sup>1</sup> and Hünefeld,<sup>2</sup> that the saliva of enraged animals, or of men in a violent fit of anger, is capable of inducing a number of highly suspicious, morbid symptoms, and more especially hydrophobia, when introduced into the blood. In the experiments made by Prinz and myself on dogs, with human saliva and the

<sup>1</sup>Physiol. der Verdauung. Würzburg, 1834, S. 28.    <sup>2</sup>Chemie u. Medicin. S. 52.

saliva of a horse, and conducted very nearly in the same manner as Wright's, excepting that we employed only filtered saliva, we never observed any symptoms of hydrophobia, even in dogs that suffered from the experiment, nor did we recognize, in the dissection of these animals, any of the pathologico-anatomical phenomena (as, for instance, in the stomach) which are usually met with in the *post-mortem* examination of mad dogs. Jacubowitsch<sup>1</sup> has also devoted the most careful attention to this subject, and instituted very accurate experiments, which not only refute Wright's statements, but expose at the same time the grounds that had led to the erroneous views arising from these experiments. The results of Jacubowitsch's experiments are as follows: human saliva does not give rise to any morbid symptoms, even when introduced in large quantities into the stomachs of dogs: unfiltered saliva produces symptoms of choking when injected into the veins: filtered saliva (which has been freed from epithelium, and other morphological substances which might obstruct the capillaries of the lesser circulation) may be injected with perfect impunity. The saliva collected during smoking contains empyreumatic substances, which give rise to symptoms of narcosis when the fluid is injected into the stomach or veins. Hertwig<sup>2</sup> has shown, by numerous experiments, that even the saliva of mad dogs, when inserted into the stomachs of other animals, or when they are inoculated with it, is unable to produce hydrophobia.

## GASTRIC JUICE.

The fluid which accumulates in the stomach after the ingestion of food, is in its pure state perfectly clear and transparent, almost entirely devoid of color, having at most but a very faint yellow tint; it has a very faint, peculiar odor, and a scarcely perceptible saline-acid taste, and is a little heavier than water; only a few morphological elements can be perceived in it; and these consist partly of unchanged cells of the gastric glands, partly of the nuclei of these cells, and partly of fine molecular matter, which is produced by the disintegration of these elements. Its reaction is very acid; it is not rendered turbid by boiling; when neutralized with alkalis a slight turbidity may sometimes be remarked. The gastric juice is distinguished from most other animal fluids by the circumstance that it remains for a very long time undecomposed, and that even when a fungous growth (mould) has appeared it always still retains its most essential character, namely, its digestive power.

The best method of obtaining gastric juice in a state of the greatest possible purity, is to feed dogs, in whom gastric fistulæ have been artificially formed, with bones which they can readily break to pieces; in the course of from 5 to 10 minutes to open the outer closed extremity of the fistula; and by means of a funnel and catheter to collect the escap-

<sup>1</sup> Op. cit. pp. 42-47.

<sup>2</sup> Beiträge zur nähern Kenntniss der Wuthkrankheit. Berlin, 1829, S. 156.

ing juice, and to separate it by filtration from flocculi of mucus, and any fragments of food that may be present. It is, however, an objection that a considerable quantity of saliva is always mixed with gastric juice obtained in this manner.

Formerly the only method of obtaining gastric juice in any available quantity was to feed animals which had been for a long time kept fasting, and to kill them in from 10 to 30 minutes afterwards. If we employ bones, tendons, or large pieces of flesh, we generally find in the stomach of the animal a gastric juice which is very suitable for the purpose of experiment, since it possesses all the properties of a normal gastric juice obtained in the preceding manner; if, however, the animals have been for a long time fasting, rather more mucus is present; this is the only difference I have ever observed. Tiedemann and Gmelin used no gastric juice in their investigations which was not collected in this manner; but in place of the above-named food they used irritant and insoluble substances (pepper-corns and pebbles).

It must be observed that this method answers very well with carnivora and omnivora, but not with herbivora (unless with ruminants); for in the latter, at all events in rabbits, we often find that after very prolonged fasting (even after the animal has died from inanition), the stomach is still full of the remains of food: in this manner, however, we never obtain a pure gastric juice, but one always containing saliva. Moreover, it is obvious that it is not a very satisfactory or useful method, since we never obtain from it more than a small quantity of gastric juice, and a large number of animals must be killed in order to obtain a sufficient quantity for the purpose of analysis or experiment.

Spallanzani, Braconnot, and Leuret and Lassaigne, obtained gastric juice without killing the animals, by making them swallow sponges attached to a string, and after some time withdrawing them from the stomach. Although these experimentalists, with the aid of this method, made many beautiful observations, and threw much light on the mysterious digestive fluid, the objections pertaining to this means are so obvious as not to require mention; the greatest being that in this way we not only collect an impure gastric juice, but that the quantity which we obtain is also very small.

The third and best method is that which we first mentioned, depending on the establishment of artificial gastric fistulæ. After Beaumont's<sup>1</sup> most admirable and decisive observations on gastric digestion, which were instituted on a man in whom a gastric fistula had become formed, in consequence of a gun-shot wound, we are in the next place indebted to Blondlot<sup>2</sup> for producing such fistulous openings in dogs. I have not found the establishment of these fistulæ by any means so easy a matter as would be inferred from Blondlot's description. A number of causes may intervene to prevent the operation from terminating favorably. Foremost amongst these, I may mention that the dogs generally bite away the ligature and the plug to which the thread passing through the

<sup>1</sup> Experiments and Observations on the Gastric Juice, and the Physiology of Digestion. Boston, 1834.

<sup>2</sup> Traité analytique de la Digestion, considérée particulièrement dans l'homme et dans les animaux vertébrés. Nancy et Paris, 1843.



stomach is fastened, and pull out the thread, so that a rupture of the stomach ensues, which is perfectly certain to cause the death of the animal; and the application of a starch bandage is seldom of any use in preventing this mischief, unless the animal be so securely tied that he cannot move himself. This cruel procedure must, further, be continued for some time, since the subsequent application of sponge plugs to dilate the fistula requires equal precautions. Hence, as far as my own experience goes, I can only recommend Bardeleben's<sup>1</sup> method of establishing such fistulæ, by which the above and many other objections to Blondlot's procedure are avoided.

According to Bardeleben, the following is the best method of proceeding. We make an incision two inches in length from the ensiform process towards the umbilicus, exactly in the linea alba; after perfectly separating the abdominal walls, we open the peritoneum for an equal length, and with two fingers seize the stomach (which, if the animal has been fed shortly before the operation, is very easily accomplished); we then form a fold about an inch in length (in which we must take care that no large bloodvessels are running), pass a ligature through it with a strong needle, and fasten the fold to a wooden peg placed transversely across the wound, which must be closed by stitches passing of course through the abdominal walls; the fold of stomach must then be included in the angle of the wound lying nearest to the navel, in order that the thread shall not cut the fold in the violent movements which accompany the vomiting that often ensues, and in which the stomach is forcibly drawn inwards. Bardeleben lays it down as a very important rule, that a doubled thread should be drawn through the abdominal muscles and fold of stomach, and that the two ends of one thread should be tied in front of the portion of stomach thus artificially prolapsed, and those of the other behind it. The wound then requires no further treatment (and this in Blondlot's method is a matter of very great difficulty); the animal's licking does no harm, since it only keeps the wound clean. The included portion of stomach soon becomes gangrenous (generally from the third to the fifth day), and is then thrown off; the fistula is then completed. To introduce a suitable canula into the fistula, which shall neither fall out nor press too hard, nor when closed shall allow fluid to escape from the stomach, is a matter of much greater difficulty. For this purpose Bardeleben has also contrived a very simple and useful apparatus, namely, a silver tube, about three-fourths of an inch long, provided at one end with a projecting border, in place of the double button-like instrument used by Blondlot: into this tube there are fitted two double hooks, united by a wire of the same length as the canula; by a well-fitting cork these hooks are so pressed upon the walls of the canula, as to render it impossible for the whole apparatus to escape from the wound. For further particulars regarding details of manipulation, I must refer to Bardeleben's memoir.

Pure filtered gastric juice contains only a small amount of solid constituents, namely, from 1.05 to 1.48%; and hence they, and especially the organic ingredients, have been very little examined.

In a specimen of human gastric juice collected by Beaumont, Berzelius found 1.27% of solid constituents; in the gastric juice of a dog

<sup>1</sup> Arch. f. phys. Heilk. Bd. 8, S. 1-7.

Blondlot found 1.00, and Leuret and Lassaigne 1.32%; and in that of a horse, Frerichs found 1.72%. I have derived the above-named numbers from experiments on the gastric juice of various dogs; it must, however, be remarked that on evaporation, the gastric juice not only loses water, but also a comparatively large quantity of hydrochloric acid, as will be seen from the experiments presently to be described. Hence it was that Tiedemann and Gmelin found that the solid constituents amounted to 1.95% in the gastric juice of a dog, to whom carbonate of lime had previously been administered, the hydrochloric acid being thus prevented from escaping, and chloride of calcium being formed.

Very different opinions have been held, up to the most recent times, regarding the nature of the free acid of the gastric juice. Prout was the first who instituted any serviceable chemical investigations regarding the gastric juice, and for a long time after the publication of his results, the presence of free hydrochloric acid in this fluid was regarded as established: it has, however, been shown (p. 93) that free lactic acid is the predominating acidifying agent in the stomach.

We have already stated all that need be said regarding the comparatively rare occurrence of hydrofluoric, acetic, and butyric acids in the gastric juice, in our remarks on those acids: we have only to add that Frerichs has recently succeeded in detecting butyric acid in the stomach of a fasting horse and of a fasting sheep, thus confirming the earlier experiments of Tiedemann and Gmelin.

In regard to the free acid in the gastric juice, I may observe that in six experiments in which I dried the gastric juice *in vacuo*, and intercepted the hydrochloric acid which was evolved (see p. 93), I found it to vary from 0.098 to 0.132%; and I then found from 0.320 to 0.583% of free lactic acid in the residue; so that if lactic acid had been the only free acid in the gastric juice (that is to say, if the acidity had depended on that acid alone) it would have ranged from 0.561 to 0.908%.

Schmidt, who analyzed specimens of gastric juice free from lactic acid, found that in nine analyses of the gastric juice (not mixed with saliva) of dogs, the free hydrochloric acid varied from 0.245 to 0.423%, the mean being 0.335%; in gastric juice containing saliva, he obtained in three analyses from 0.1708 to 0.3353%, the mean being 0.2337%; while the gastric juice from the fourth stomach of the sheep yielded as the mean of two analyses 0.1234%. The gastric juice of the sheep always contained a little lactic acid, which, however, was apparently not secreted in the glands in the walls of the stomach, but formed by fermentation from starch.

It is not at all improbable that the quantity of free acid in the gastric juice is as variable as that of the alkali in the saliva; any one, however, who has occupied himself with experiments of this nature, will see that these numbers can only give an approximative idea regarding the quantity of acid in the gastric juice; for, independently of the circumstance that the fluid collected from a gastric fistula is never obtained in a state of entire purity, the methods adopted for exciting the flow of the secretion exert an essential influence on its constitution. The gastric juice used in my experiments was collected from three dogs at very different times, after they had fasted for twelve hours. I gave them

fatty bones, and collected the gastric juice in from 10 to 25 minutes afterwards; it was only by the repetition of the process that I could gradually collect a quantity of gastric juice sufficient for analysis, so that each of the six determinations may be regarded as giving a mean result. I determined the whole amount of the free acid of the filtered gastric juice, by saturating it with carbonate of baryta, and then calculating the quantity of free lactic acid from the sulphate of baryta precipitated by sulphuric acid.

Blondlot, by some erroneous process, was induced to believe that gastric juice did not decompose carbonate of lime, and was hence led to conclude that the acid reaction of the gastric juice depended solely on the acid phosphate of lime; Dumas, Melsens, and Bernard have found that not only the carbonate but also the basic phosphate of lime is soluble in gastric juice, as also are even zinc and iron, hydrogen being simultaneously developed—properties which a solution of acid phosphate of lime does not possess.

In addition to lactic acid, the solid residue of the gastric juice contains an extraordinary quantity of *metallic chlorides*, namely, chloride of sodium with smaller quantities of the chlorides of calcium and magnesium, and traces of protochloride of iron.

Schmidt, moreover, found that *chloride of ammonium* was constantly present in the gastric juice; its quantity varied in the pure gastric juice of the dog from 0.0372 to 0.065% (the mean being 0.047%), and in the gastric juice mixed with saliva from 0.0276 to 0.084%, while in the gastric juice of the sheep it averaged 0.0475%. The gastric juice (free from saliva) of the dog contains on an average 0.4256% of *fixed chlorides*, while that which is mixed with the saliva contains 0.588%, the addition of the saliva to the gastric juice inducing an augmentation of the chlorides of sodium and calcium; of the latter Schmidt found only 0.0624% in pure gastric juice, while the quantity amounted to 0.1661% in the mixed fluid. The gastric juice (containing saliva) of the sheep contains on an average 0.6% of fixed chlorides.

On evaporating gastric juice we obtain a residue consisting of crystals of chloride of sodium, moistened with a yellowish syrupy mass, which consists principally of lactate of soda. The presence of protochloride of iron may almost always be easily recognized in strongly evaporated gastric juice by means of ferridcyanide of potassium.

*Phosphate of lime* is only present in small quantities in filtered gastric juice.

When the juice abounds in mucus or cells, this salt usually occurs in larger quantities, if we estimate it by the ash left by the residuc.

*Alkaline sulphates and phosphates* cannot be detected in pure gastric juice; neither can *ammoniacal salts*.

For if ammonia were present, on evaporating fresh gastric juice over hydrated magnesia, and extracting the residue with alcohol, there would either be a development of ammonia (which is not observed), or hydrochlorate and lactate of ammonia would be found in the alcoholic solution; on precipitating the bases from this fluid with sulphuric acid, we find no trace of ammonia in the deposit. If, however, the ammonia were combined with the magnesia as triple phosphate, this might be readily

discovered by a microscopic examination of the residue insoluble in alcohol; for even when this residue was treated with a little phosphoric acid and hydrochloric acid (perfectly free from ammonia), and the solution digested with magnesia, the triple phosphate was not formed. I can, therefore, only believe that the hydrochlorate of ammonia found by Braconnot, Tiedemann and Gmelin, and others, must have been formed during the chemical examination by the action of free hydrochloric acid on mucus or some other nitrogenous animal substance.\*

In addition to the mineral constituents, we also find in the gastric juice certain organic substances which, however, in consequence of the extremely small quantities in which they occur, have been very little examined; these are a substance soluble in water and in absolute alcohol (formerly known as osmazome), and a substance soluble only in water, and more or less perfectly precipitable by alcohol, tannic acid, corrosive sublimate, and the salts of lead; the latter, which seems to be a mixture of several different substances, constitutes the true digestive principle; its solution, on being boiled, certainly loses the property of effecting the characteristic change on the protein-bodies and gelatinous substances, but does not coagulate like albumen, as was formerly supposed from experiments performed with artificial gastric juice.

With regard to the *ferment* of the gastric juice, Schmidt obtained it by neutralizing the fluid with lime-water, evaporating to the consistence of oil, and precipitating with anhydrous alcohol; this precipitate was then dissolved in water, and thrown down with bichloride of mercury; in the organic matter of this mercury-compound Schmidt found 53·9% of carbon, 6·7% of hydrogen, and 17·8% of nitrogen.

The mean amount of the *organic matters*, both in pure and in mixed gastric juice, was a little above 1·7%, while the mineral constituents averaged 1·0%.

Schmidt found no essential difference in the composition of the gastric juice of dogs, after they had been fed for a long time solely with vegetables, or solely with flesh. It cannot, however, be a mere accidental coincidence, that the highest numbers for the phosphate of iron were found in four cases after the use of vegetable food.

The ratio in which the mineral constituents of the gastric juice stand to the organic, is a somewhat varying one; in the gastric juice of the horse, Gmelin found 1·05% of organic, and 0·55% of inorganic constituents, while on the other hand Frerichs found 0·98% of organic, and 0·74% of inorganic matters; in the gastric juice of the dog Frerichs found 0·72% of organic, and 0·43% of inorganic constituents; while I found from 0·86 to 0·99% of the former, and from 0·38 to 0·56% of the latter.

By the term *artificial gastric juice* we understand a fluid which is obtained by treating the glandular tissue of the stomach in a peculiar manner with dilute hydrochloric acid, and which possesses the characteristic property, in common with the natural gastric juice, of converting nitrogenous articles of food into soluble, non-coagulable substances.

After Eberle<sup>1</sup> had shown that the gastric juice, when removed from the animal body, retains the property of inducing peculiar changes in the food, and that by digesting the mucous membrane of the stomach

<sup>1</sup> Physiologie der Verdauung. Würzburg, 1834.

with extremely dilute acids, we obtain a fluid which possesses true digestive powers, it was proved by Schwann<sup>1</sup> that it is only the glandular structure of the stomach which possesses the property of yielding a digestive mixture with acids, and further, that corrosive sublimate throws down a precipitate from it, which possesses the digestive power in a high degree. To this substance Schwann gave the name of *pepsin*. Wasmann,<sup>2</sup> who investigated the subject even more fully than Schwann, demonstrated that the source of the gastric juice and of this pepsin lay in the gastric glands, which he carefully observed and described; he likewise attempted to exhibit pepsin in a purer state.

He proceeded in the following manner: the glandular layer in the stomach of the pig, which extends chiefly from the greater curvature towards the cardia, was carefully detached and washed, without being cut up; then digested with distilled water at a temperature of from 30° to 35°. After some hours the fluid was poured away, the membrane was again washed in cold water, and then digested in the cold with about six ounces of distilled water, and repeatedly washed, till a putrid odor began to be developed. The filtered fluid was transparent, viscid, and without any reaction; it was now precipitated with acetate of lead or corrosive sublimate; the precipitate was carefully washed and decomposed with sulphuretted hydrogen; the pepsin was then precipitated by alcohol from the watery solution in white flocks.

The pepsin thus obtained, forms, when dry, a yellow, gummy, slightly hygroscopic mass; in its moist state it is white and bulky; it dissolves readily in water, and always retains a little free acid so as to redden litmus; it is precipitated by alcohol from its watery solution; mineral acids induce a turbidity in a solution of neutralized pepsin, which disappears on the addition of a small excess of the acid; but if there be a considerable excess of the acid, there is a flocculent deposit; it is only imperfectly precipitated by metallic salts, and not at all by ferrocyanide of potassium; it has been asserted that pepsin is coagulated by boiling, but Frerichs has shown that the coagulation is merely dependent on its admixture with albumen.

This substance possesses the converting power in so high a degree, that, according to Wasmann, a solution containing only one-sixtieth thousand part, if slightly acidulated, dissolves coagulated albumen in six or eight hours. This property of pepsin is not destroyed by alcohol; and in this respect Wasmann and Schwann coincide: it is, however, lost when the solution is boiled or carefully neutralized with potash; in both cases the fluid becomes turbid.

Almost simultaneously with Wasmann, similar experiments on the digestive principle were made by Pappenheim<sup>3</sup> and Valentin,<sup>4</sup> and subsequently by Elsässer,<sup>5</sup> with artificial gastric juice; and they all arrived at the same general results; but pepsin sufficiently pure for chemical analysis has never been exhibited up to the present time.

<sup>1</sup> Pogg. Ann. Bd. 38, S. 358.

<sup>2</sup> De digestionem nonnulla, diss. inaug. Berol. 1839.

<sup>3</sup> Zur Kenntniss der Verdauung. Breslau, 1839.

<sup>4</sup> Valentin's Repert. Bd. 1, S. 46.

<sup>5</sup> Magenerweichung der Säuglinge. Stuttgart, 1846. S. 68 ff.

As in Wasmann's method of procedure, putrid parts and digested particles of food were always mixed with the artificial gastric juice, I struck upon the following method of obtaining a digestive fluid in a state of the greatest possible purity.

The stomach of a recently killed pig having been properly cleaned, I detached from it the portion of mucous membrane in which the gastric glands chiefly lie.

As this piece of mucous membrane still contains a tolerably thick layer of submucous areolar tissue, or of the so-called vascular coat, in which the gastric glands are in a manner imbedded, this cannot be at once employed in the preparation of the digestive fluid, since then a quantity of digested gelatinous substance would be mixed with it. This source of error cannot be entirely avoided, since in every mode of treatment heterogeneous elements of tissue will be mixed with the glandular contents. In order, however, to obtain the latter in as pure a state as possible, the piece of mucous membrane, after lying for an hour or two in distilled water at the ordinary temperature, must be gently scraped with a blunt knife or spatula; the pale grayish-red, tenacious mucus which adheres to the blade must be placed in distilled water, and the mixture must be kept at the ordinary temperature for two or three hours, being frequently shaken in the interval: a little free acid must then be added, and the mixture placed for half an hour or an hour in a hatching-oven at a temperature of from  $35^{\circ}$  to  $38^{\circ}$ . By this time, the fluid will be found to have lost much of its viscosity, and it is now only slightly turbid; it passes readily through the filter, in the form of a perfectly limpid fluid with a scarcely perceptible yellow tint.

These and similar artificial mixtures are of much service, as experience has indeed fully shown, in the investigation of different conditions and phenomena in relation to digestion; but they are far less suited than gastric juice discharged from the living animal for experiments having for their object to isolate as much as possible from the unessential ingredients, and to render fit for chemical analysis, the true digestive principle, or the group of substances which constitute it. If the gastric juice from the living animal be always mixed with a little saliva, that fluid interferes far less with an accurate analysis than the albumen and the different peptones in the artificial digestive fluids; and even if we could separate the albumen, the peptones would still be associated with the digestive principle, as indeed they are even with the natural gastric juice, although in a far less degree. Notwithstanding the labors of many observers, it appears by no means impossible that by repeated investigations we may so limit the digestive principle as to find a chemical expression for it, whether we can exhibit the actual substance or not. Frerichs, in his classical article on digestion, has hit upon the right line of investigation—upon the only course which can lead to definite limits—when he precipitated the natural gastric juice with alcohol; unless too much alcohol be added, the greater part of the peptones, and also of the aqueous extractive matter of the saliva, remains in solution, as indeed does a little pepsin. The precipitate dissolves pretty freely in water, from which it is precipitated by corrosive sublimate, protochloride of tin,

basic acetate of lead, and tannic acid, and in an imperfect manner by neutral acetate of lead; it does not become turbid on boiling, exhibits strong digestive properties when treated with dilute hydrochloric or with lactic acid, but, like the gastric juice, is deprived of them by boiling, by absolute alcohol, and by neutralization with alkalies; in an alkaline solution it very soon becomes putrid, and in a neutral one it seems to give rise to the formation of fungi; but when rendered acid, it remains for a very long time without suffering decomposition, exactly as natural gastric juice. Frerichs has proved that the flocks precipitated by alcohol contain sulphur and nitrogen.

We shall discuss the true sources of the pepsin, the gastric glands, and their contents, in the histologico-chemical part of this work.

C. Schmidt<sup>1</sup> has propounded a very interesting view regarding the nature of the digestive principle; he regards it as a conjugated acid, whose negative constituent is hydrochloric acid, with Wasmann's non-acid or coagulated pepsin as an adjunct, and assumes that it possesses the property of entering into soluble combinations with albumen, gluten, chondrin, &c.; according to him, it more nearly resembles ligno-sulphuric acid than any other conjugated acid, and as this becomes disintegrated into dextrin and sulphuric acid, so the *pepsin-hydrochloric acid* becomes separated at 100° into Wasmann's coagulated pepsin and hydrochloric acid, and in either case it is equally impossible to reproduce the conjugated acid from its proximate elements after their separation. On bringing the complex acid in contact with an alkali, the adjunct—the substance which has been in combination with the hydrochloric acid—is precipitated. Schmidt believes that he has ascertained that an artificial digestive mixture which has expended its solvent and digestive powers, regains them on the addition of free acid; and that when hydrochloric acid is added, the pepsin-hydrochloric acid is expelled from its combination with albumen, &c., and thus regains its former properties, while the newly added hydrochloric acid enters into its well-known soluble combinations with albumen, &c. By the repeated addition of hydrochloric acid, a digestive fluid or this pepsin-hydrochloric acid might preserve its digesting power forever, unless the fluid became saturated with the dissolved substances, or the conjugated acid underwent decomposition.

Ingenious as this view of Schmidt's undoubtedly is, and singularly as it seems to harmonize with certain facts, there are other and very important facts which appear to render its correctness doubtful. The existence of this pepsin-hydrochloric acid has not been recognized by any analysis of a combination of it with a mineral base or with an albuminous substance. Although I have instituted numerous experiments regarding the quantitative relations between the digestive fluid and the substances to be digested, I cannot ascertain that there are any such proportions between the acid and the digested substance as at all accord with the ordinary acid or basic combinations of acid and base; and further, the digested substances (the peptones) separated by the acid, are altogether different from the original albumen, fibrin, casein, &c.,

<sup>1</sup> *De digestionis natura*, etc. Diss. inaug. Dorp. Liv. 1846; and *Ann. d. Ch. u. Pharm.* Bd. 61, S. 22-24

which, however, according to Schmidt, combine in a simple manner with this complex acid, and then directly undergo solution. Further grounds for opposing this hypothesis will become apparent when we enter more fully into the consideration of the peptones.

Very little is known regarding the *abnormal constituents* which, under certain physiological or pathological conditions, may occur in the gastric juice. We know that in the normal condition, the stomach, when it is empty, is invested with a layer of mucus, which exhibits no reaction with vegetable colors. In gastric catarrh, this *mucus* accumulates in larger quantities, and on chemical examination is found to present little difference from the secretions of other mucous membranes; and, like them, it only in a slight degree possesses digestive powers on the addition of a free acid: even while in the stomach it appears in part to undergo decomposition, and subsequently, on being mixed with amylaceous or saccharine food, to enter into abnormal processes of fermentation, as, for instance, *acetic*, *butyric*, and *lactic fermentation*. The contents of the stomach then contain far more free acid than occurs in them in normal digestion. The two last-named processes of fermentation are especially promoted by the presence of fat, which gives rise to heartburn, a sensation of constriction in the throat, and vomiting; and at the same time, there is often a revulsory (antiperistaltic) motion of the intestinal tube, which causes a regurgitation of bile into the stomach, and this is an additional impediment to digestion. *Biliary matters* cannot, however, strictly speaking, be regarded as abnormal constituents of the gastric juice, since they are never produced from the same sources as that secretion; they are, however, so frequently met with, that I have made few examinations of human bodies, or even of recently killed healthy animals, in which I have not discovered biliary constituents in the contents of the stomach lying near the pyloric end.

The contents of the stomach, in *post-mortem* examinations, and sometimes also the matters which are vomited in cases of gastric catarrh, are perfectly neutral or even alkaline on their outer surface, which is turned towards the walls of the stomach, while the inner parts often exhibit a very strong acid reaction; this phenomenon, wonderful as it appears at first sight, is obviously dependent on the circumstance that there must simultaneously have been a deficient secretion of gastric juice, and such slight movements of the stomach as not to have sufficiently mixed the contents with one another; and hence, either that the inner portions have undergone one of the above-mentioned acid fermentations, or that they have retained the acid reaction peculiar to the food.

It appears as if heterogeneous matters in the animal body made a repeated circulation through the gastric glands, as they seem to do through the salivary glands, before they are removed by the kidneys, or undergo change in any other part; at least this seems to be shown by the experiments of Bernard,<sup>1</sup> who injected solutions of sulphocyanide of potassium and of perchloride of iron into different veins of the same dog, and first observed the formation of sulphocyanide of iron in the gastric juice.

<sup>1</sup> Arch. gén. de Méd. 4 Sér. T. xi. p. 310.



It is universally known that in uræmia, or after extirpation of the kidneys, *urea* is secreted by the gastric glands.

Since the time of Nysten (see p. 154), *urea* has often been found in the vomited matters in cases of uræmia, consequent on Bright's disease or on cholera. Bernard and Barreswil<sup>1</sup> have made two interesting experiments in reference to this point. After extirpating the kidneys in dogs, they found that at the commencement of the retention of the urinary constituents in the blood, the gastric juice contained no *urea*, but a very large quantity of hydrochlorate of ammonia, without, however, being less acid than in the normal state: it is worthy of notice that the gastric juice, under these circumstances, was secreted very copiously without the irritation caused by the presence of food, that is to say, when the dog was fasting. As long as the gastric juice remained acid, these chemists found no *urea* in the blood; they found it, however, as soon as well-marked morbid symptoms were established in the animal; and in this case there was only a little gastric juice, which, moreover, was secreted in a decidedly alkaline state, and contained much carbonate of ammonia.

In two cases in which I analyzed vomited matters, I found *urea* when the patients presented none of the phenomena of uræmia; the vomited matter had a distinctly urinous odor, and, moreover, contained uric acid. It was afterwards proved that the patients, who were hysterical girls, had been drinking their own urine, and had simulated retention of that excretion. Rayer<sup>2</sup> has recorded a similar case. In accordance with Bernard's experiments, we very often find carbonate of ammonia in vomited matters, and especially in the contents of the stomach after death. In the group of symptoms which are associated with cholera and Bright's disease, and to which we give the name *uræmia*, I have always found the contents of the stomach and the vomited matters strongly alkaline, and always rich in carbonate of ammonia, but never containing *urea*. The symptoms indicating uræmia must, however, have their foundation in something more than in the mere decomposition of *urea* into carbonate of ammonia; for when I injected dilute solutions of carbonate of ammonia in various proportions into the blood-vessels of cats and dogs, convulsions, and even tetanic spasms (in the case of large doses) ensued, which, as is well known, do not pertain to the ordinary phenomena of uræmia, while vomiting did not occur in either class of animals, although it may be induced readily in both. The stomach was usually only slightly reddened, and presented no essential change in relation to its amount of mucus.

We are as yet unable to make any decisive statement regarding the quantity of gastric juice secreted in twenty-four hours; indeed, on this point, we are at present entirely devoid of data. We only know that, in the healthy state, its secretion is entirely dependent on the ingestion of food, and that some articles of diet excite a more copious effusion of gastric juice than others. Thus, for instance, sugar, aromatic substances, spirit of wine, and alkalies, when introduced into the stomach, immediately excite an almost overflowing secretion of gastric juice; while, on the other hand, animal substances, which remain for a longer

<sup>1</sup> Arch. gén. de Méd. 4 Sér. T. xiii. p. 449-465.

<sup>2</sup> Maladies des Reins, p. 285.

period in the stomach, require a far greater quantity of gastric juice for their perfect conversion.

According to my experiments, 100 grammes of the fresh gastric juice of a dog cannot, on an average, effect the solution of more than 5 grammes of coagulated albumen (calculated as dry). Now if we assume that an adult man receives into the stomach about 100 grammes of albuminous matter in twenty-four hours, there must be secreted 2000 grammes, or 4 pounds of gastric juice, for the digestion of this quantity. In order to determine the *daily quantity of gastric juice which is secreted*, Bidder and Schmidt employed dogs with gastric fistulæ, which they made to lie on the left side when the secretion commenced, by which means the passage of any part of the gastric juice through the pylorus into the duodenum was prevented: the secretion was, moreover, collected on various days (with a considerable interval between them) and at different periods after the last meal. They collected 823 grammes of gastric juice from a dog weighing 16 kilogrammes, which on the whole was submitted to 14 observations, extending over 12 hours. (These observations of course not being continuous.) Another dog weighing 12 kilogrammes yielded 231 grammes in 4 hours (it having been submitted to 6 observations). In the first case there were 103, and in the second, 115 grammes of gastric juice secreted for each kilogramme's weight of the animal. We may, therefore, assume that the dog yields, at the least, 10% of its weight of gastric juice in 24 hours. We further know that in the healthy state the secretion of gastric juice is for the most part dependent on the ingestion of food, and that some kinds of food excite a more copious flow of this fluid than others. There are some substances, such, for instance, as sugar, aromatics, spirits, and alkalies, which when introduced into the stomach, immediately excite an almost gushing secretion of gastric juice, while other substances, as for instance, animal food, require a far larger quantity of gastric juice for their metamorphosis, in consequence of the much longer time of their retention in the stomach. As we shall return to this subject in our remarks on the processes of digestion and nutrition, we need not enter into it more fully in the present place.

After the preceding observations, there can be no doubt regarding the *physiological function* of the gastric juice. The gastric juice serves not merely to dissolve, but also to modify the nitrogenous elements of food, as, for instance, the protein-compounds and their derivatives. It was formerly believed that its only use was to convert insoluble and coagulated substances into the corresponding soluble matters, and thus to render them capable of resorption, and that it did not in any way affect the soluble substances. If we have since convinced ourselves that the casein is first coagulated by the gastric juice, in order again to be converted by it into a soluble substance, we yet believe that soluble albumen neither requires nor undergoes any such alterations in order to be resorbed, or, as we commonly express it, to be assimilated (Tiedemann and Gmelin). On the other hand, we learn, from a positive experimental inquiry, what are the products which are developed during the process of digestion; and we ascertain that, by the action of natural or artificial gastric juice on protein-bodies or gelatigenous matters, there

are formed thoroughly new substances, which, although they coincide in their chemical composition and in many of their physical properties, with the substances from which they are derived, essentially differ from them, not only in their ready solubility (in water, and even in dilute alcohol), but in having now lost the faculty of forming insoluble combinations with most metallic salts. The formation of these substances, which we designate as *peptones*, depends solely on the action of the gastric juice, and occurs without the evolution or absorption of any gas, and without the production of any secondary substance.

Beaumont was the first who observed that non-coagulated albumen also undergoes a change on the stomach, while Tiedemann and Gmelin, and subsequently Blondlot, believed that they had arrived at the opposite conclusion, from their experiments. Any one may, however, very easily convince himself that the blood-serum and the albumen of eggs when stirred with water and filtered, are rendered as strongly turbid by the gastric juice as by any other dilute acid; the gastric juice, whether it be natural or artificial, often exerts little or no further influence on the albumen, when its digestive power is gone in consequence of the partial or total loss of the free acid. If, however, we again add fresh acid, we perceive the gradual conversion of the albumen in the diminution of the quantity of the coagulable substance, unless, like Blondlot, we use too small a quantity of gastric juice for the albumen; finally, the fluid ceases to give any trace of ordinary albumen, either when boiled, or on the addition of nitric acid or of any other test. The same process may be observed in natural digestion. If, for instance, we observe the contents of the stomachs of dogs with a gastric fistula, after they have swallowed such solutions of albumen, we find that the contents at first have only a slight acid reaction (whether they are clear, or turbid from the partial precipitation of the albumen, is a point which cannot be decided, in consequence of the invariable presence of mucus). Very soon, however, after from 5 to 10 minutes, so much gastric juice has been secreted, that the alkali of the soluble albumen is not merely saturated, but the whole digestive mixture has assumed a strong acid reaction. Here also we may observe a gradual diminution of the coagulable matter; but I will not venture to deny that a part of the albumen may pass in an uncoagulated condition into the small intestine, in a state of health, as has been observed by Tiedemann and Gmelin. Mialhe<sup>1</sup> has also convinced himself of the metamorphosis of soluble albumen during gastric digestion. In relation to the products of the digestion of soluble albumen, I have been unable, with the means we at present possess of analyzing such complex bodies as the protein-compounds, to discover any difference between the peptones of soluble and coagulated albumen.

Schwann, in accordance with the ideas and nomenclature of that day, gave the names of *osmazome* and *ptyalin* to the substances which resulted from the digestion of albumen; Mialhe was the first to discover that a single, easily soluble substance, is produced from the digestion of albumen or other protein-bodies, and gave to it the name of *albuminose*. We shall return, in a future part of the work, to the properties of albumen-peptone.

<sup>1</sup> Journ. de Pharm. et de Chim. 3 Sér. T. 10, p. 161-167.

The fibrin of the blood is not dissolved by the gastric juice in the same manner as by a solution of nitre (see p. 313), but it is converted into a non-coagulable, soluble substance, *fibrin-peptone*.

That soluble casein is coagulated in the stomach before it undergoes the actual process of digestion, has been long known; it being proved by observing milk which has been vomited, and by the well-known property of the calf's stomach (rennet) to induce coagulation. More recent observations have only shown that the casein thus coagulated requires in general a longer time for its solution than most other protein-bodies, and that here also as in the other bodies of this class, the more easy or difficult digestibility principally depends on the atomic grouping in which it is secreted; hence, according to Elsässer,<sup>1</sup> the casein of woman's milk, which only coagulates into a sort of jelly, is more easily digested than the clotted and more firmly coagulated casein of cow's milk.

Globulin, vitellin, legumin, and other protein-bodies, behave, according to my experiments, both in natural and artificial digestive fluids, precisely the same as albumen.

It is singular that gluten, chondrin, and gelatigenous tissues, during their digestion in the stomach, are converted into substances which, in their physical and in most of their chemical properties, perfectly correspond with the peptones of the protein-bodies. The degree of the solubility of these substances is however essentially dependent on mechanical relations; actually formed gelatine is more readily changed than areolar (cellular) tissue, and the latter far more quickly than tendon and cartilage; indeed, as a general rule, the latter do not remain in the stomach sufficiently long to be completely digested, but for the most part are carried away undigested with the excrements.

We shall treat of the digestibility of mixed food and of the individual animal tissues, when we consider the digestive process generally.

Very little attention has hitherto been paid even to the best-known peptones; indeed, until Mialhe published his researches, positively nothing was known regarding their physical or chemical relations. This chemist erroneously regarded the soluble substances produced by digestion from the protein-bodies and from the gelatigenous tissues as perfectly identical. The following properties, which Mialhe attributes to his albuminose, are certainly correctly observed, and are common to most of the peptones; in the solid state the digested substances are white or of a pale yellow color, possess little taste or odor, and dissolve readily in water and slightly in spirit, but not at all in absolute alcohol. The watery solutions of these substances are not precipitated by boiling, by acids, or by alkalies, but deposits are thrown down by metallic salts, by chlorine, and by tannic acid.

My own observations lead me to the belief that all the peptones are white, amorphous bodies, devoid of any odor, and having merely a mucous taste, soluble in every proportion in water, and insoluble in alcohol of 83%; their watery solutions redden litmus; they combine readily with bases—with alkalies as well as with earths—so as to form neutral salts, which are very soluble in water. The aqueous solutions of these salts, are only precipitated by tannic acid, corrosive sublimate, and

<sup>1</sup> Die Magenerweichung der Säuglinge. Stutt. u. Tüb. 1846.

if caustic ammonia has been previously added, by acetate of lead; all other metallic salts, even nitrate of silver and alum, produce no precipitate, and even basic acetate of lead only induces a slight turbidity, which disappears on the addition of an excess of the test. No precipitation or turbidity is produced by the addition of mineral or organic acids, either in a concentrated or in a very dilute state; even chromic acid fails to produce any appreciable effect. The ferrocyanide and ferridcyanide of potassium, when added to solutions acidified with acetic acid, occasion only a slight turbidity.

I have been unable to obtain the peptones perfectly free from mineral substances: I have, however, obtained them free from phosphates and hydrochlorates, so that their ash contained only alkaline carbonates or carbonate of lime, with small quantities of alkaline sulphates. With regard to the quantity of sulphur in the peptones, I found it to be constantly the same as that in the substances from which they were derived; thus, for example, in the peptone of the albumen of eggs, after deducting the alkali or lime, I found in three experiments, 1.579, 1.659, and 1.600% of sulphur, the mean being 1.602%, which coincides almost to the very decimal places with Mulder's determination of the amount of sulphur in the albumen of the egg. This sulphur appears, however, to be contained in the peptone, in precisely the same form as it exists in the albumen; at all events, when treated with alkalis it yields very distinct indications of sulphur, both with the salts of lead and with silver-foil. In my repeated analyses I have been unable to detect any differences between the quantities of nitrogen, carbon, and oxygen, contained in the peptone and in the substance from which it was derived, nor can I infer from my quantitative results, that the conversion of the protein-bodies into peptones is accompanied by an assimilation of water, as might have been supposed. The metamorphosis may be appropriately compared with that of starch into sugar, or even better perhaps, with that of cholic (Strecker's cholalic) acid into choloidic acid.

I have prepared the peptones either from the natural gastric juice of dogs or from artificial digestive fluid obtained from the pepsin-glands of the stomach of the pig and from coagulated albumen, fibrin, casein, legumin, gluten, and chondrin in a state of extreme purity, by allowing them to remain in contact at the necessary, somewhat elevated temperature, till the greater part of the substance to be digested had dissolved; the whole mixture was then boiled and filtered; the acid fluid was somewhat evaporated over carbonate of lime, and after a second filtration, was concentrated to the consistence of honey. The addition of alcohol (of 83%) precipitated the lime-and-peptone compound, but dissolved the chloride of calcium; the undissolved portion, which was very hygroscopic on exposure to the air, and soon ran into a varnish-like mass, was now boiled with absolute alcohol, and was finally extracted, while still hot, with ether containing alcohol. The alkali-compound admitted of being easily prepared from the lime-compound by means of alkaline carbonates. The peptones were obtained nearly, but not perfectly, free from mineral constituents, by carefully removing the baryta, or a great part of it, from their baryta-compounds, by means of sulphuric acid.

The alkaline carbonates only partially remove lime from the lime-pep-

tone, but they entirely free it from phosphate of lime; if, for instance, the alkaline peptone-solution after being freed by filtration from the carbonate of lime thrown down by carbonate of potash, be slightly acidified with acetic acid, evaporated and freed from acetates by extraction with alcohol, neither carbonate of soda nor ammonia produces any precipitate when added to the aqueous solution, but a precipitate is caused by oxalate of ammonia; the ash consists here almost entirely of carbonate of lime. Thus albumen-peptone contains, for instance, 5.53% of lime. Hence the saturating capacity of albumen-peptone = 1.67, and its atomic weight = 5960.

I have obtained perfectly similar results in analyzing other peptones, the details of which I shall describe more fully in another part of this work; this much, however, may be concluded with certainty, that the digestion of the protein-compounds is something more than a simple formation of the well-known hydrochlorate of albumen, as was formerly supposed, and as has partly been assumed by Schmidt, in the hypothesis to which we have previously referred.

The following facts are worthy of notice in reference to the *digestive power* of the gastric juice: it is *suspended* by boiling, by saturating the free acid with an alkali or even with phosphate of lime, by sulphurous, arsenious, and tannic acids, by alum and by most metallic salts; and it is very much *impeded* by the addition of alkaline salts, or by saturating the fluid with peptones or other organic substances, either nitrogenous or non-nitrogenous. The addition of *water* to a gastric juice which has been already saturated by a peptone, enables it to digest an additional quantity of protein-substances; the digestive power is also restored to a certain degree by the repeated addition of *free acid*. Too much free acid without due dilution with water, entirely suspends the digestive power. The most favorable ratio of the free acid of the gastric juice is when 100 parts of the latter are saturated by about 1.25 of potash. Hydrochloric and lactic acids are the only acids which yield energetic, active digestive fluids with pepsin; sulphuric, nitric, and acetic acids yield with pepsin a digestive mixture of only slight power; while phosphoric, oxalic, tartaric, and succinic acids can in no degree replace the lactic or hydrochloric acid in the process of digestion. Fats, when added in certain quantities to the gastric juice, promote the conversion of the protein-compounds into peptones.

It need excite no wonder that sulphurous, arsenious, or tannic acid should suspend the digestive power of the gastric juice, for it is well known that these substances check other metamorphoses, and especially the phenomena of fermentation. Taking into consideration the chemical properties of pepsin, and its power of combining with metallic salts and other substances, we could hardly expect that the above-named substances would exert any other effect on the digestive power of the gastric juice.

Wasmann has very clearly shown that no digestion is possible unless the gastric juice contains a free acid; indeed he was led to the view that the digestive power resides "in solo acido." This latter view is, however, sufficiently controverted by the experiments of numerous observers; we need, for instance, only refer to those of Blondlot, who believed that he had

shown that the peptones are bodies which are essentially different from the soluble hydrochlorates and lactates of the protein-compounds. The simplest experiment is indeed sufficient to show that dilute acids are incapable of producing the same effects as the gastric juice.

It was shown by Elsässer<sup>1</sup> that a digestive mixture which had been already saturated with a digested substance, and had consequently lost its digestive powers, regains them in part, either by being diluted with water, or by the addition of free acid. Different views have been founded on these experiments (as, for instance, by Elsässer and Schmidt), but it appears to me that questions of this nature can only be decided by quantitative determinations; and I<sup>2</sup> have instituted many series of experiments in reference to this point, without, however, as yet succeeding in obtaining a definite formula for these relations, which could be expressed in numbers.

Elsässer, from his experiments with an artificial digestive fluid, concludes that from 3 to 4% of hydrochloric acid,  $\text{HCl.HO}$  (and, therefore, probably from 1.2 to 1.6% of the anhydrous acid,  $\text{HCl}$ ), is the most favorable ratio; besides this, the quantity of solid constituents in it should not exceed 1.25%.

Wasmann, and other observers, for the most part ascribe the peptic force to the free acid in general. My numerous experiments have, however, led me to the result which I have already mentioned, namely, that other acids when associated with pepsin, possess only a slight digestive power, and that even hydrochloric acid, in which phosphate of lime had been dissolved to the saturating point, no longer possesses any digestive force when united with pepsin.

Very different views were formerly deduced from the results of positive investigation, in reference to the activity of the alkaline chlorides in digestion. I, myself,<sup>3</sup> formerly believed that I had ascertained that the addition of chloride of sodium to the gastric juice, promoted the solution of the protein-bodies, but more recent and extensive experiments have convinced me that every kind of neutral alkaline salt very much impedes the digestive process.

It is easy to demonstrate,<sup>4</sup> by experiments on living animals, and with both artificial and natural gastric juice, that fat very much promotes the conversion of the protein-bodies into peptones. This observation has been confirmed by Elsässer.

It further follows from the numerous experiments of Bidder and Schmidt, that pure gastric juice considerably exceeds gastric juice mixed with saliva in its digestive power,—a fact obviously dependent on a portion of the free acid of the gastric juice being saturated by the alkaline saliva. They likewise found that the addition of bile to the gastric juice entirely suspends its solvent action, although the mixture still exhibits a decided acid reaction.

This latter experiment distinctly explains how it is that, when still undigested albuminous matters pass into the intestine, the gastric juice loses all power over them. If there be an acid reaction in the duodenum, this does not depend upon the presence of free hydrochloric acid, but on

<sup>1</sup> Op. cit.

<sup>3</sup> Op. cit.

<sup>2</sup> Ber. der Ak. der Wiss. z. Leipz. 1849, S. 8–50.

<sup>4</sup> Simon's Beiträge. Bd. 1, S. 22.

that of the biliary acids isolated by it. Since these are either very readily resorbed or else are insoluble, we commonly fail to observe an acid reaction in the jejunum even after the use of a flesh-diet.

With regard to the *quantities of albuminous substance* which can be dissolved by definite quantities of gastric juice, I have found that 100 grammes of the fresh gastric juice of the dog are able, on an average, to dissolve 5 grammes of coagulated albumen (this being the mean of eight experiments, in which the extremes were 6.14 and 4.317 grammes). Schmidt, who instituted similar experiments, arrived at a far lower result: as a mean of 27 experiments, he found that 100 grammes of gastric juice dissolved only 2.2 grammes of albumen, the highest number which Schmidt found being 3.95 grammes. The method which I pursued in these investigations was the same as that which I adopted in my experiments with artificial gastric juice. The higher numbers which I obtained were probably dependent on the presence of lactic acid in the fresh gastric juice, while Schmidt only operated on gastric juice, in which there was no lactic acid. Since many conditions favoring the solution of the protein-bodies co-operate within the stomach, and since the gastric juice obtained from fistulous openings, probably possesses less digestive power than that which is secreted from uninjured stomachs, Bidder and Schmidt very correctly infer that the gastric juice may be able to dissolve a larger amount of albuminous matter than the results of our experiments would seem to show.

Now, if we know the quantity of the gastric juice which is secreted in twenty-four hours (see p. 449), and the quantity of albumen which is dissolved by a definite quantity of gastric juice, we can readily ascertain the quantity of albumen which can be daily digested in the stomach. Since a dog secretes about 100 grammes of gastric juice for every kilogramme's weight of its body, that animal would only be able to digest 5% of its weight of albumen (reckoned as dry). But it appears from the numerous experiments of Schmidt, that a dog, in order to keep in condition on an exclusive flesh-diet, should take for every kilogramme's weight of its body, 50 grammes of flesh containing 10 grammes of dry albuminates; hence the gastric juice secreted by the dog would only suffice for the digestion of half of the albuminates necessary for the nutrition of the dog,—a result which, paradoxical as it may appear in connection with the preceding view regarding the digestion of albumen, stands in the most perfect accordance with other observations presently to be described.

The experiments of most observers, agree in showing that the gastric juice exerts no perceptible action on the ordinary *non-nitrogenous foods*. The fats may certainly, as we have already mentioned, exert an influence on the gastric digestion, but they undergo no recognizable chemical change. Starch, gum, and sugar, when placed in pure gastric juice at the temperature of the animal body, do not undergo any change corresponding to the digestion of nitrogenous bodies. We shall return to the consideration of mixed and natural vegetable food, when we treat of the process of digestion.

If the fats exert an influence on digestion, we can hardly conceive that this action is due to mere contact, and that it is unaccompanied by



any change in the fat itself, but the quantity of fat which acts and is modified in this way in the digestive process, is so minute as not to be appreciable in our analyses; it is evident that it is not in the stomach that the fats are digested.

We have already mentioned (see p. 436) that Bernard believed that he had discovered that acid saliva, like acid gastric juice, digests animal food, and that alkaline gastric juice, like alkaline saliva, digests starch; this view is, however, opposed by the positive experiments of Mialhe and Jacobowitsch. I have also convinced myself that neither natural or artificial gastric juice, even when rendered strongly alkaline, exerts any action on starch. According to Jacobowitsch, saliva mixed with gastric juice converts starch into sugar; and I have confirmed this experiment with acid, neutral, and alkaline mixtures.

As the vegetable substances are permeated by saliva and gastric juice, we find that they are softened and partially loosened in their texture in the stomach; the gastric juice here naturally exerts its digestive power only on their nitrogenous constituents, while the non-nitrogenous materials probably only undergo a preparation in the stomach for the changes which are normally effected in the small intestines.

Pure gastric juice antagonizes the ordinary processes of fermentation, and hence lactic, acetic, and alcoholic fermentation are excluded from the sphere of gastric digestion, so long as this process is a normal or physiological one. At the very most only a part of the cane or milk sugar introduced into the stomach, can be converted into glucose.

[Gruenewaldt<sup>1</sup> and Schroeder<sup>2</sup> have recently published excellent Theses on the human gastric juice, the former taking up its physical and chemical characters, and the latter investigating its digestive powers. Their observations were conducted at Dorpat, under the superintendence of Bidder and Schmidt, on an Esthonian peasant, Catharine Kütt, in whom there was a gastric fistula (the origin of which they could not ascertain) in the left side, at the lower border of the mammary gland, between the cartilages of the ninth and tenth ribs.

The following are the most important results of Gruenewaldt's observations:

For every kilogramme of bodily weight there are 264 grammes of gastric juice secreted in the 24 hours, the mean daily quantity of gastric juice secreted by this woman being 14.016 kilogrammes, or about 31 lbs., a quantity somewhat larger than that deduced by Schmidt from his experiments on dogs.

The *sarcina* was frequently observed in this fluid, obtained from the fistula, both when the stomach was empty and when full, the woman being apparently in perfect health. Hence Gruenewaldt agrees with Virchow,<sup>3</sup> that this organism must not be regarded as a special symptom of a peculiar form of disease.

In relation to the chemistry of this fluid, he found that, when obtained from the empty stomach, it was never acid, but always neutral or slightly alkaline. He gives the particulars of three analyses which were made

<sup>1</sup> *Succi gastrici humani Indoles physica et chemica*, etc. Dorp. Liv. 1858.

<sup>2</sup> *Succi gastrici humani Vis digestiva*, etc. Dorp. Liv. 1853.

<sup>3</sup> *Arch. f. pathol. Anat.* Bd. 1, S. 268.

by Schmidt. In all these cases the secreted fluid was of a very pale reddish tint, moderately acid, formed coagula when boiled, and gave indications of the presence of much sugar, by Trommer's test. When heated it yielded the odor of butyric and metacetic acid. The spec. grav. of the first specimen was 1.020.

	I.	• II.	III.
Water, . . . . .	954.134	961.251	954.401
Solid constituents, . . . . .	45.866	38.749	45.599
An albuminate coagulating at 100° C. (Pepsin), . . . . .	0.780	} 31.939	} 38.659
Sugar, albuminates not coagulating by heat, lactic, and butyric acids, and ammonia, . . . . .	38.430		
Chloride of potassium, . . . . .	0.704	} 6.810	} 6.940
Chloride of sodium, . . . . .	4.263		
Potash (in combination with the organic acids), . . . . .	0.179		
Phosphate of lime, . . . . .	1.030		
Phosphate of magnesia, . . . . .	0.470		
Phosphate of iron, . . . . .	0.010		

He proves by experiments which are fully described in his Thesis, that the acid which is liberated on the application of heat, consists of much butyric acid, with a little metacetic and, probably, acetic acid; and that the human gastric juice contains *no free hydrochloric acid*. He regards the butyric and lactic acids as products of the metamorphosis of the carbo-hydrates; and, finally, he is persuaded that the acid reaction of the gastric juice, when mixed with food, owes its origin to the organic acids, which are contained in or developed from that food.

Schroeder's Thesis is divided into three sections. In the first, he considers the action of the human gastric juice on amylaceous matters; in the second, its action on the albuminates, and especially on flesh; and in the third, he briefly notices the part which it takes in the metamorphosis of matter. The only point especially deserving of notice is the description of the analyses of the gastric juice of the same woman, obtained unmixed with food, by irritating the gastric mucous membrane of the empty stomach with pepsin. An acid, clear gastric juice was then obtained, containing free hydrochloric acid; it would thus appear, that in Gruenewaldt's experiments, this acid had been neutralized by the alkali of the saliva.—G. E. D.]

## BILE.

The bile of different animals does not present exactly the same physical properties; in the following points, however, we find a tolerable identity of character between the different kinds of this secretion. When

derived from the gall-bladder, the bile occurs as a mucous, transparent fluid, capable of being drawn out in threads, of a green or brown color, of a bitter but not astringent taste and sometimes leaving a rather sweet after-taste, and of a peculiar odor, which, when the bile is warmed, often vividly reminds the observer of musk. Its specific gravity is about 1.02: bile does not diffuse itself readily through water, unless the mixture be stirred; it is usually weakly alkaline, often perfectly neutral, and only in disease, and then rarely, acid. Bile in its ordinary state, before its mucus is removed, putrefies very readily, but when it is freed from mucus, putrefaction is not easily induced.

Fresh human bile can only be obtained from the bodies of criminals immediately after their execution; the bile of animals is commonly obtained from the gall-bladder immediately after they have been killed; in the case of animals like the stag and the roe, which possess no gall-bladder, it is only rarely that we can obtain from the larger biliary ducts a quantity of bile sufficient for an accurate analysis. With the view of more accurately studying the relations of the biliary secretion and its influence on digestion, Blondlot,<sup>1</sup> Schwann,<sup>2</sup> and C. Schmidt<sup>3</sup> have established biliary fistulæ in animals. These fistulæ are made in the same way as gastric fistulæ, by cutting through the abdominal walls, but the incision in this case must be somewhat longer; we then raise up the lower border of the left lobe of the liver, and search for the ductus choledochus at the point where it opens into the duodenum; if the animal has a gall-bladder, the best plan is to tie the above-named duct at two spots and to cut away some of the intervening portion; all the bile must then flow through the cystic duct into the gall-bladder. The latter must then be separated as well as possible, and as far as is necessary, from its attachment to the liver, and drawn forth from the abdominal cavity, while any prolapsed intestine must be returned, and the wound, as when we establish a gastric fistula, closed by strong twisted sutures; an incision must then be made into the gall-bladder, and the outer edges of the wound secured, as in the case of the stomach. After this operation, which is far the more severe of the two, animals much more frequently die from peritonitis and enteritis, than after the establishment of a gastric fistula. If we make the ductus choledochus open directly on the external surface of the body, the prognosis is still more unfavorable, since the canal becomes attached with less facility and certainty to the abdominal walls. It is advisable to introduce a small glass tube or a silver canula into the duct immediately after the operation, in order to prevent the bile from coming in contact with the lips of the wound.

Physiologists have ever held the most different and opposite views regarding the function of the liver and of its secretion, and even at the present day the subject is involved in the greatest obscurity; and in regard to the nature of the bile itself, since zoo-chemical analyses of it were first attempted, there have been so many difficulties and impediments in the way of prosecuting them, that it is only during the last few years that any light has been thrown upon this most obscure of all the

<sup>1</sup> *Essai sur les fonctions du foie et de ses annexes.* Paris, 1846.

<sup>2</sup> *Müller's Archiv.* 1844. S. 127-162.

<sup>3</sup> *Buchheim's Beitr. z. Arzneimittellehre.* Leipz. 1849, S. 116.

departments of animal chemistry. The most distinguished chemists of our time, founding their views on the most exact experiments, have been led to perfectly different results regarding the constitution of the bile; we shall, however, consider it in the following manner, which is based on the most recent investigations conducted under Liebig's auspices, and explains many of the former points of difference:

Every kind of bile contains two essential constituents, namely, a resinous and a coloring constituent.

The *resinous constituent* is, as a general rule, the soda-salt of one of the *conjugated acids* described pp. 201–208, whose adjunct is *glycine* or *taurine*.

The *coloring principle* of the bile has also been described pp. 277–283: it occurs in combination with an alkali in the bile.

A third never-failing constituent is the *cholesterin*, described pp. 244–248.

Besides these essential constituents, we also find *fats* and combinations of the alkalies with *fatty acids* in the bile.

Moreover, we find in the bile the same *mineral salts* which occur in most other animal fluids; namely, chloride of sodium (the principal salt), a little phosphate and carbonate of soda, phosphate of lime and magnesia, and extremely minute quantities of iron and manganese, but no alkaline sulphates. No salts of ammonia are found in fresh healthy bile. The relation of the potash and soda in the bile of different animals—a fact noticed by Bensch, but more prominently evolved by Strecker—is deserving of careful attention: the bile of salt-water fishes contains almost exclusively potash-salts, while that of the herbivorous mammalia contains almost exclusively soda-salts; whereas from the nature of the food of the animals, we should have expected to have met with the opposite result. We have already alluded to the presence of copper in the bile. (See p. 403.)

Finally, a greater or lesser quantity of *mucus* always occurs in the bile. This, like other varieties of mucus, is mixed with numbers of epithelial cells; here, however, the mucus-juice very much preponderates over the epithelium.

Fresh normal bile contains no morphological elements except the cells of cylindrical epithelium thrown off from the mucous membrane of the biliary ducts and the gall-bladder; these cells often remain grouped together in their natural arrangement.

It is needless to introduce in this place any historical sketch of the manifold experiments and views which have been adduced in reference to the composition of the bile, since they are described with more or less minuteness in all our text-books of Animal Chemistry, and in every monograph on the bile, and since they do not throw the slightest light on the complex nature of this fluid and of its constituents. In reference to the writings of Berzelius, it seems, however, necessary to state, that according to his view, which has very recently been defended by Mulder, the most essential constituent of the bile is not an acid in combination with soda, but an indifferent substance named *bilin*, which in its decomposition gives rise to those substances which were formerly described by Gmelin, and subsequently by Demarçay and others, as occurring in the

bile. Any one who carefully studies the chemical characters of taurocholic acid (the choleic acid of Strecker), and compares them with those which are ascribed by Berzelius to his bilin, will readily detect the causes of the error by which that chemist was led to assume the existence of an indifferent bilin; and they will no longer wonder that all who have repeated the positive experiments of Berzelius have quite as thoroughly confirmed them, as those which were instituted by Liebig and his pupils, but led them to a different view of the subject.

If these disputes amongst the first chemists of our time, regarding the constitution of the bile, should at the first glance cause the faith of the physician in the extreme certainty of chemical investigation to stagger, and should blight his hopes of our ever attaining to an exact humoral pathology, let him carefully study the grounds of these differences of opinion, and he will be convinced that he has no cause for doubting the accuracy and certainty of chemical inquiries. He must especially bear in mind that different chemists have entered upon the study of this complex fluid from different points of view, without, as it were, meeting one another half-way, and thus obtaining a general survey; further, it must be recollected that the bile undergoes decomposition with extraordinary rapidity, and scarcely any chemist assumes that he has employed perfectly undecomposed bile in his analyses; indeed it has even been believed that the decomposition of the bile commences within the healthy living body in the gall-bladder. Moreover it is clear that by employing different methods of analysis, we obtain different products of metamorphosis. Finally, we must always recollect that the comprehension of the results of the analysis—the consideration of the objects perceived—is always subjective, that is to say, it is the result of an intellectual process. Hence we see that even where all the facts were fully confirmed, none of the opinions that have been expressed regarding them have preponderated, since none of them could be made to harmonize with all the results of different experimenters. This object has, however, been attained, as we have already observed, by means of Strecker's experiments, conducted under the auspices of Liebig, although, as might be expected, there still remain some few obscure points requiring further elucidation.

In reference to the resinous acids of the bile, we have little to add to what we have already communicated regarding Strecker's investigations. Mulder,<sup>1</sup> however, still defends the opinion of Berzelius, that bilin is secreted by the liver, but believes that it undergoes a complete decomposition in the gall-bladder. Strecker,<sup>2</sup> on the other hand, has extended his investigations, and has analyzed the bile of various classes of animals; and hitherto he has found that the only difference in the composition of the bile of different animals is in the varying proportions in which the taurocholic and glycocholic acids (the choleic and cholic acids of Strecker) exist in them. In the bile of fishes (*Gadus morrhua*, *Pleuronectes maximus*, *Esox lucius*, *Perca fluviatilis*), Strecker found that the resinous constituents consisted almost entirely of alkaline taurocholates, with mere traces of alkaline glycocholates; and, singularly enough, that the

<sup>1</sup> Scheik. Onderz. D. 5, p. 1-104.

<sup>2</sup> Ann. de Ch. u. Pharm. Bd. 70, S. 149-198.

potash-salts preponderated in the salt-water, and the soda-salts in the fresh-water fishes. The researches of Strecker show that the bile of the dog contains almost exclusively taurocholate of soda, and a similar fact had been previously ascertained by Schlieper in reference to serpents' bile. It seems, moreover, to follow from Strecker's experiments, that the nature of the food (in the case of dogs) exercises no influence on the composition of the bile. The bile of the sheep contains, according to Strecker, a mixture of much taurocholate of soda with a comparatively small amount of glycocholate. The bile of the goose, according to Marsson's investigations, contains almost exclusively taurocholic acid. Hyocholic acid has only been found in the bile of the pig; on the other hand, it appears that the small quantity of sulphur formerly detected by Strecker and Bensch in the bile of the pig depends on the presence of a hyocholic acid; that is to say, that besides the glycine-yielding hyocholic acid (glyco-hyocholic acid), there also occurs a taurine-yielding acid in very small quantity—an acid in which the taurine is united with the same resinous acid ( $C_{50}H_{40}O_8$ , the hyocholic acid of Strecker) with which glycine is combined in hyocholic acid. The products of the decomposition of hyocholic acid have been carefully studied by Strecker. It is worthy of notice that this chemist, in examining pigs' bile from which the biliary acids had been removed by hydrochloric acid, discovered a very strong sulphurous base, which is capable of combining even with carbonic acid.

The peculiar pigment has never yet been found to be absent in the bile of any animal. In the bile of carnivorous and omnivorous animals, including man, we have a brown pigment, the cholepyrrhin of Berzelius; while in the bile of birds, fishes and amphibia, we usually find an intense green pigment, biliverdin. The brown bile-pigment is moreover never contained in a state of freedom, but is always in combination either with soda or lime; in the latter case it is insoluble, and may be easily recognized in the brown granules which we sometimes observe in examining the bile with the microscope. A microscopico-chemical analysis affords a ready proof that these granules consist of the combination of cholepyrrhin with lime.

The quantitative relations of the biliary constituents have not as yet been very accurately investigated; the following statements may, however, be regarded as representing with tolerable accuracy the mean composition of the bile.

Normal *human bile* contains, according to the determinations of Freichs,<sup>1</sup> about 14%, or a little more of solid constituents; ox-bile, from 10 to 13%; and pigs' bile (according to Gundelach and Strecker<sup>2</sup>) from 10.6 to 11.8%: the amount of water may, however, be as variable in the bile as in most other animal secretions.

Gorup-Besanez<sup>3</sup> found 9.13% of solid constituents in the bile of an old man, and 17.19% in that of a child aged twelve years; but whether the bile is always more diluted in old age than in childhood, is a question that must be decided by further investigations.

The organic constituents of human bile amount to about 87% of the

<sup>1</sup> Hannov. Ann. Bd. 5, II. 1 u. 2.

<sup>2</sup> Ann. d. Ch. u. Pharm. Bd. 62, S. 205-232.

<sup>3</sup> Untersuch. über die Galle. Erlangen, 1846, S. 44.

whole solid residue ; and much the same ratio seems to obtain in the bile of animals.

Berzelius obtained 12·7% of ash from the residue of ox-bile ; and Bensch<sup>1</sup> 13·15% from that of calves' bile, 11·86% from that of sheep's bile, 13·21% from that of goats' bile, 13·6% from that of pigs' bile, 12·71% from that of foxes' bile, 10·99% from that of fowls' bile, and 14·11% from that of the bile of fresh-water fishes.

The alkaline taurocholates and glycocholates constitute by far the greater part of the organic constituents, and amount to at least 75% of the whole of the solid constituents of the bile.

The investigations of Bensch and Strecker show that the bile of most of the animals included as yet in their experiments, contains a preponderating quantity of taurocholate of soda. As taurocholate of soda ( $\text{NaO.C}_{52}\text{H}_{41}\text{NO}_{13}\text{S}$ ) contains 6% of sulphur, we may readily estimate the taurocholic acid contained in any quantity of bile, from the amount of sulphur contained in the portion soluble only in alcohol. Schlieper<sup>2</sup> found 6·2% of sulphur in purified serpents' bile, that is to say, in its alcoholic extract ; in that of a dog, Bensch found 6·2%, but Strecker only 5·9% ; in that of a fox, Bensch found 5·96% ; while in that of the sheep, Strecker found from 5·7 to 5·3%. Hence we perceive that the bile of these animals contains taurocholic acid almost exclusively, while ox-bile, whose alcoholic extract contains only 3% of sulphur, contains taurocholic and glycocholic acids in nearly equal proportions. As the bile of the pig contains only from 0·3 to 0·4% of sulphur, we may hence draw our conclusions regarding the small quantity of hyocholic or taurohyocholic acid contained in it.

No correct determinations have been made regarding the amount of the pigment, the cholesterin, or the fats and fatty acids in the bile.

Moreover, the quantitative determinations of the mineral constituents of the bile, cannot be regarded as altogether trustworthy ; the only established fact seems to be that there is a quantity of soda or potash present which is equivalent to the resinous acids ; but the pigment and the fatty acids are also combined with alkalis ; ordinary analyses of the ash, and even those made in accordance with Rose's directions, do not by any means lead to the result that all the alkali combined with organic matter has been accurately determined. The ash of ox-bile is almost the only one which has been carefully examined ; it contains, according to Weidenbusch,<sup>3</sup> 27·70% of chloride of sodium, and about 16% of tribasic phosphate of soda, with only 3·025% of basic phosphate of lime, 1·52% of basic phosphate of magnesia, 0·23% of peroxide of iron, and 0·36% of silica.

I have convinced myself that the bile—at all events, that ox-bile—contains preformed *alkaline carbonates*, by the same experimental proof which I adopted to demonstrate the presence of salts in fresh blood. If we place bile under the receiver of an air-pump, and abstract the air till the fluid appears to boil, and if we then add acetic acid to the bile thus freed from gas, and again form a vacuum around it, very large quantities of carbonic acid will be evolved, even with the first strokes of the pump.

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 65, S. 215.

<sup>2</sup> Ibid. Bd. 60, S. 109.

<sup>3</sup> Pogg. Ann. Bd. 76, S. 886.

I must here remark that, in performing this experiment, perfectly fresh bile, from which the mucus had been removed by alcohol, was employed; and this, on the addition of acetic acid, yielded no precipitate of fine granules which might have facilitated the formation of vesicles of aqueous vapor. The experiment may also be easily performed by simultaneously placing acidified and non-acid bile *in vacuo*, when the difference more readily strikes the eye. In 100 parts of fresh ox-bile, I found in two quantitative determinations, 0.0846, and 0.1124 parts of the simple carbonate of soda.

We must here further remark, in connection with the uncertainty of ash-analyses, that the soda combined in the bile with organic substances, will appear in the ash as carbonate of soda; this, however, occurs only in extremely small quantity, for the greater part of the soda is saturated by the sulphuric acid which is formed during the combustion of the taurocholic acid and the mucus. The sulphuric acid in the ash, resulting from this source, is, however, extremely variable, according to the mode in which the incineration has been conducted. Weidenbusch, who employed Rose's method for the determination of the ash, satisfied himself that, even in this way, a great part of the sulphur contained in these organic substances has volatilized, and therefore does not appear in the ash as sulphuric acid. The researches of all the more recent chemists show that fresh bile contains scarcely a trace of sulphuric acid. A portion, however, of the soda which was in combination with organic substances, is found in the ash as phosphate of soda, a salt which—as ordinary phosphate of soda ( $2\text{NaO} \cdot \text{HIO} \cdot \text{PO}_5$ )—most probably exists preformed in the bile. Thus it is easy to see that, under certain conditions, even no carbonate of soda may be found in the ash.

In normal human bile, Frerichs found from 0.20 to 0.25% of chloride of sodium, and an equal quantity of phosphate of soda. Theyer and Schlosser found 3.56% of this salt in the bile of the ox.

The determination of the *mucus* in normal bile is not to be depended on, for the bile which has been examined has usually been expressed from the gall-bladder with such force that a large quantity of the epithelium, from the lining membrane of that organ, becomes mixed with the secretion. In ox-bile I found only 0.134, and in human bile 0.158% of mucus, when I used every precaution to avoid this source of error.

Very little is known regarding the changes which the bile undergoes under purely physiological conditions. When it has been retained for a long time in the bladder, as, for instance, in cases of prolonged fasting, it is concentrated. A highly nitrogenous diet not only increases the biliary secretion, but likewise renders it more concentrated than ordinary bile.

The differences in the physical characters and in the composition of freshly secreted bile and of bile that has been retained for a long time in the gall-bladder, have been successfully investigated by Bidder and Schmidt. The fresh bile of carnivorous animals (dogs, cats, crows) varies from a yellow to a yellowish-brown color; while in herbivorous animals (rabbits, sheep, geese) it is green; the color of the *cystic bile* in animals whose *hepatic bile* is yellow or brown, has, however, always



more or less of a tendency to green, and when the animals have not fed for 20 hours or more is of a deep green; while, if examined  $2\frac{1}{2}$  or 3 hours after feeding, it is of as light a yellow or yellowish-brown color as the hepatic bile. Since the hepatic bile gradually becomes green on exposure to the air, and the yellow tint may be again restored by deoxidizing agents, there can be no doubt that this change of color depends on oxidation, and that the bile retained in the gall-bladder is impregnated by the circulating blood with so much oxygen as to induce this altered color.

The prolonged retention of the bile in the gall-bladder induces, however, not only a partial oxidation of this secretion, but also a strong concentration—a fact which has been established by the numerous observations of Bidder and Schmidt, and has been confirmed by Nasse. The former inquirers found that the fresh hepatic secretion of cats, dogs, and sheep contained on an average 5% of solid constituents, which in the case of cats and dogs rose to 10 or even 20% in the cystic bile, according to the duration of its retention in the gall-bladder: in sheep, on the other hand, the amount only rose to 8%; in rabbits, whose fresh bile contains only 2% of solid constituents, the amount in the cystic bile may rise to 15%. The fresh bile of geese and crows contains about 7% of solid matters, which in the cystic bile of the former may rise to 20%, and in that of the latter even to 25%. The bile, therefore, restores to the blood and lymph a greater or smaller quantity of water, according to the duration of its retention in the gall-bladder.

As in the case of other secretions and excretions, *heterogeneous constituents* may find their way into the bile. The older writers have often asserted that the bile contained *albumen*, but they doubtless mistook mucus for albumen. Albumen is, however, sometimes found in the bile, especially in fatty liver (although rarely), in Bright's disease, and in the embryonic state. In a five months' human embryo I found no true bile in the gall-bladder, but only yellow-colored albumen and mucus.

Thénard has observed and described a peculiar form of albuminous bile, which was perfectly colorless; it occurred in certain cases of fatty liver. Frerichs directs attention to the film which is often formed on the surface of morbid bile during evaporation; but this membrane may be formed by the coagulation of mucus-juice as readily as by casein-like substances; in two cases of fatty liver I believe, however, that I actually found albumen, for I treated the bile with acetic acid as long as any precipitate (consisting of mucus, and biliary and fatty acids) continued to be thrown down, and then boiled the filtered fluid with hydrochlorate of ammonia (see p. 298); a coagulum was then formed, which yielded the ordinary reactions of the protein bodies. Bernard<sup>1</sup> was the first who detected albumen in the bile in Bright's disease. When the bile contains pus, as is sometimes the case in abscesses of the liver, albumen must obviously be present.

In the case of obliteration of the cystic duct, in consequence of which *hydrops vesicæ felleæ* (as it has been termed) was developed, I found that

Bouisson, de la bile, de ses variétés physiologiques et de ses altérations morbides. Montpellier, 1842.

the colorless fluid in the gall-bladder contained traces of coagulable matter, in addition to epithelium, and mucus-juice.

The occurrence of *urea* in the bile after extirpation of the kidneys, has been already noticed (see p. 154); this substance has also been found in the bile in Bright's disease and in cholera.

The alcoholic extract of the bile of a man who died with the symptoms of fatty degeneration of the kidneys, was extracted with aqueous ether; the ethereal extract, when treated with nitric acid, yielded most distinct crystals of nitrate of urea; fat-globules were also present in it. Stan-  
nius and Sthamer<sup>1</sup> failed in detecting urea either in the bile or in the kidneys of animals whose kidneys had been extirpated.

Bizio once discovered a dark red, non-bitter bile in a patient suffering from icterus; it contained an *emerald-green pigment*, to which he gave the name of *erythrogen*, from its volatilizing at 40° and giving off a red vapor.

I found a similar substance in a case of acute yellow atrophy of the liver; its behavior was precisely the same as that of Bizio's erythrogen; it was insoluble in water and ether, partially soluble in alcohol, but dissolved readily in concentrated mineral acids without any change of color. I obtained it, like Bizio, by diluting the bile with water; the insoluble portion was boiled with water, upon which a fatty green mass separated on the surface, which had the above-named properties in common with erythrogen.

In the bile of a child who died suddenly, I<sup>2</sup> found a considerable quantity of *sulphide of ammonium*.

It is sufficiently obvious that this *sulphide of ammonium* had not been separated from the blood by the liver; the only singularity is, that it should have been found in such large quantity in the bile, when the examination was made sixteen hours after death. Unfortunately nothing was known of the previous history of the case.

With the exception of the above-mentioned changes, the only other alterations in morbid bile (which can obviously only be obtained from the body after death), are of a quantitative nature in reference to the individual constituents, or are represented by modifications of the pigment. The bile has been found to be poor in solid constituents in persons who have died from severe inflammatory affections, especially from pneumonia, and likewise in fatal cases of dropsy; it is even more aqueous and attenuated in certain cases of typhus; and in diabetes there is always an excess of water. In tuberculosis the bile is very frequently, although not invariably, poor in solid constituents.

In cases of tuberculosis, Gorup-Besanez usually found the bile of the ordinary consistence; but Frerichs always found it attenuated, unless when the tuberculosis was complicated with fatty liver. This difference may be readily accounted for; Frerichs probably analyzed bile in cases in which an anæmic condition had been induced, in consequence of abundant effusion (as, for instance, where diarrhœa had been excited by intestinal ulceration, or where there had been pleural or peritoneal dropsy; his last case was one of obsolete tubercle). Again, no one who has exa-

<sup>1</sup> Arch. f. phys. Heilk. Bd. 9, S. 201-219.

<sup>2</sup> Schmidt's Jahrbücher der ges. Med. Bd. 25, S. 16.

mined the blood of tuberculous patients before and after the exudation has been thrown off, can wonder that the bile should present a thin liquid appearance after an attack of acute tuberculosis. In tuberculosis combined with fatty liver, Frerichs, like Gorup-Besanez, found the bile dense, since in this condition the blood is less poor in solid constituents, and the hepatic affection is itself opposed to a copious secretion of dilute bile. Both chemists found the bile very diluted and scanty in typhus; the bodies from which the bile was obtained, were those of persons in whom the morbid process was already localized, or when death was induced, as it frequently is, not directly by the typhus, but by the subsequent anæmia. In two cases of typhus, in which the *plaques* in the intestine were only just recognizable, I found the bile dense; and every pathological anatomist must recollect cases in which the bile was tough and consistent, and, therefore, rich in solid constituents in persons who had died from typhus. In every case Frerichs found from 93 to 96% of water in the bile, and Gorup-Besanez for the most part a somewhat smaller quantity.

The *solid constituents* are commonly *increased* in those abdominal diseases in which the motion of the blood in the larger veins is impeded, and where, as in certain cases of heart-disease, the blood accumulates in excessive quantity in the portal vein and the hepatic vessels. The motion of the blood in the hepatic capillaries is (as we know from physiological researches) so torpid, that if there be any impediment, as, for instance, disease of the heart, to the passage of the blood in the *vena cava*, and any check to the escape of the blood from the liver through the hepatic veins, an almost entire stagnation of the blood-current in the liver must ensue.

In cholera we also find the bile dense, tough, and consistent; this condition is likewise due for the most part to mechanical conditions; the blood of cholera patients is so tenacious and thick, that even in the vicinity of the heart it moves slowly, and thus causes a disturbed state of the circulation generally; and this effect is the more striking in the hepatic circulation, when, moreover, in consequence of the blood being deficient in water, a less aqueous bile must be secreted.

The *mucus* is often relatively increased when the bile is very dilute; indeed, in typhus we sometimes find little else in the gall-bladder than mucus, the resinous constituents being almost entirely or altogether absent; and the same is observed in catarrh of the biliary ducts.

In the absence of quantitative determinations, we cannot decide whether the separation of *crystals of cholesterin*, which we can sometimes observe with the microscope in morbid bile, is associated with an absolute augmentation of this lipid. Gorup-Besanez has only occasionally observed this phenomenon in very concentrated bile.

*Free fat* is always present in the bile, but is held in solution by the taurocholic acid; occasionally, however, in examining morbid bile by means of the microscope, we may detect fat-globules, which we must be careful not to confound with the globules of separated biliary acids, which are often observable. Gorup has found fat-globules in the bile of persons who had died from typhus and from tuberculosis (in the colliquative stage). • We have already alluded to the fact (see p. 227), that in such cases free fat also passes into the urine.

It is very seldom that the bile has been found to have an *acid* reaction, and in none of these cases has it been carefully analyzed.

Solon, Scharlau, and Gorup-Besanez occasionally found the bile acid in typhus; this may, however, depend partly on the spontaneous decomposition of the bile, and the consequent liberation of its resinous acids, and partly on the fact that pus is effused into the gall-bladder; for, as we shall subsequently show, this fluid, when contained in an enclosed space, often becomes acid with great rapidity.

According to Solon, the bile is sometimes as acid as chlorine, and *bleaches litmus*. I believe that I have observed two cases of the kind which probably led Solon to adopt this view; this bile certainly decolorized litmus paper, so that it remained neither blue nor red, but its coloring matter was so dissolved out, or covered by the yellow pigment of the bile, that the original tint seemed wholly to have disappeared; in a less degree this is the case with every specimen of bile.

The following may be regarded as the simplest method of *analyzing the bile*. We treat the fluid with half its volume, or from that to its own volume, of spirit (83%). This generally only throws down mucus, which carries with it any epithelium that may be present; we rinse the precipitate first with spirit, and then with water, and dry and weigh it. The bile thus freed from mucus, is deprived of its water, by being placed first on the water-bath, and, subsequently, under the air-pump on a sand-bath heated to 100°; the high temperature in this process of desiccation is less necessary for the purpose of effecting the drying quickly than for converting the residue of the bile by the rapid evaporation of the water into a porous, spongy, puffy substance, which admits of being extracted by the ordinary menstrua with comparative facility. As the residue of scarcely any other animal fluid attracts moisture so readily from the air, especial care must be paid to the weighing of its solid constituents; after the mass has cooled *in vacuo*, air from which the aqueous vapor has been extracted by chloride of calcium must be drawn into the receiver, and the weighing must be completed as quickly as possible. The residue must then be extracted with anhydrous ether, a process requiring much time, because we cannot pulverize it like the residues of other animal fluids, in order to submit to further analysis a newly-dried and weighed quantity. The ethereal extract contains fat, and not unfrequently also a little of the resinous biliary matters, which may be separated from the fat by aqueous spirit. The residue insoluble in ether, which contains the essential constituents of the bile, must be dissolved in absolute alcohol; we must then remove the greater part of the alcohol by distillation or evaporation, and treat the concentrated fluid with ether as long as any turbidity is observable: there then generally only remains a very little alkali in combination with a fatty acid, and some chloride of sodium in the ethereo-alcoholic fluid: the fluid with its precipitate must, however, stand for a considerable time in a cool place, because the alkaline glycocholate only separates very slowly. The salts of the biliary acids which are thus separated, are unfortunately always mixed with bile-pigment, from which they can only rarely be separated by the addition of chloride of calcium to their alcoholic solution (namely, when the pigment consists of true cholepyrrhin). By dis-

solving in alcohol a portion of the mixed glycocholates and taurocholates precipitated by ether, and by adding sulphuric acid to the solution, we can determine the quantity of the soda or potash in combination with these salts and with pigment, and we can ascertain whether or not ammonia be present. Unfortunately the determination of the alkali in this way is not strictly accurate, since a little chloride of sodium and soda in combination with a fatty acid, always occur in the precipitate thrown down by the ether, and thus contribute their alkali to that with which the biliary acids were combined. An exact separation of the taurocholic and glycocholic acids is impossible (as indeed is obvious, from what has been stated in p. 209); consequently the best method of calculating the amount of the biliary acid yielding taurine, is by determining the quantity of sulphur in the biliary salts<sup>1</sup> which have been precipitated by ether; for this purpose we oxidize a weighed portion of them with potash or soda and nitre in the dry way, and determine the sulphuric acid that is formed. As, even if sulphates had been present, no sulphuric acid could have got into the alcoholic extract, it is obvious that all the sulphuric acid that is found, must have been derived from the sulphur which was in combination with organic matter; and taurocholic acid is the only sulphurous substance contained in the alcoholic extract.

The residue of the bile insoluble in absolute alcohol must now be determined with the view of checking the analysis; it contains pigment, partly free and partly in combination with lime, alkaline and earthy phosphates, with a little alkaline carbonate and chloride of sodium, very rarely sulphate of potash, but often a little taurine; its amount is generally so small that any further quantitative determination, as, for instance, by means of diluted spirit, water, acids, &c., is hardly practicable.

Those who have studied all that has been stated in the earlier part of this volume regarding these substances and their properties, need hardly be informed that the methods of analyzing the bile can, and indeed must, be variously modified, and that the method we have just given can only serve the purpose of an illustrative scheme.

We have also stated all that is necessary regarding the quantitative determination of the cholesterin and fatty acids. These substances can only be determined quantitatively when a very large quantity of bile is submitted to analysis.

*Biliary concretions* must be ranked amongst the morbid products of the secretion of the liver. Few points in pathological chemistry, in the earlier period of that science, have received so much attention as gall-stones; but all the very numerous observations which have been made regarding them are reducible to the following facts: these concretions occur principally in the gall-bladder, more rarely in the biliary ducts; in women more frequently than in men, and especially in aged persons: they often co-exist with cancer of the liver or of other organs, but it cannot be positively affirmed that carcinoma is a predisposing cause of gall-stones, since both these adventitious products specially pertain to advanced age and to the female sex; each is, however, often found in-

<sup>1</sup> [The alkaline taurocholates and glycocholates.—G. E. D.]

dependently of the other. Gall-stones appear to be of more common occurrence in England, Hanover, and Hungary, than in other countries. Most gall-stones are so rich in cholesterin, that the other constituents are of very secondary importance; all, however, contain one or more nuclei, consisting of traces of mucus and earthy phosphates, but principally of an insoluble combination of lime with bile-pigment: a large number of gall-stones are formed of a mixture of *cholesterin* and *pigment-lime*;<sup>1</sup> the latter is sometimes uniformly distributed through the concretion, in other cases we observe alternating layers of cholesterin and the brown pigment, and in others again we find only a little cholesterin in the dark-brown mass of pigment-lime.

There is a third kind of concretion which is comparatively rare, namely, the black or dark-green variety; this contains another modification of the pigment, which, however, in this case also is combined with lime: this variety is usually free from, or at all events very poor in cholesterin.

Biliary concretions, in which carbonate and phosphate of lime are the principal ingredients, are very rare. (Bailly and Henry Steinberg).

It is singular that *uric acid* has occasionally been found in gall-stones (Stöckhard,<sup>2</sup> Marchand).<sup>3</sup>

All gall-stones absorb a little bile, which may be readily abstracted from the pulverized concretion with water or cold alcohol.

The *forms* of gall-stones are extremely varied: while some are very regular and symmetrical, others assume the most unaccountable shapes.

Bramson<sup>4</sup> has undoubtedly indicated an important point in relation to the formation of the majority of gall-stones—namely, that it depends on the separation of a compound of pigment with lime.

Although Bramson's view has been much contested, we can undoubtedly recognize the presence of a compound of pigment with lime in the residue of the nuclei both of cholesterin concretions and of the brown gall-stones after extraction with alcohol and water, although we are, as yet, unable to establish a definite proportion between the pigment and the base. Every residue which is rich in pigment always contains a greater or lesser quantity of earthy phosphates and a little mucus; these earthy phosphates most probably originate from the mucus, which, however, like the protein-bodies in the formation of phlebolites, gradually dissolves and disappears; for the phosphates never stand in a constant ratio to the mucus remaining in the concretion; the mucus may also contain a little lime, which on incineration is converted into carbonate and sulphate; moreover, we sometimes meet with oxalate of lime, although only in very small quantity; I have never found preformed carbonate of lime in the brown residue of gall-stones (if present, it may be very readily detected by observing, under the microscope, the effect produced by a little acid on the substance previously moistened with water and freed from all air-bubbles). Sulphate of lime does not exist preformed, or at all events it is only present in very small quantity.

The ratio of the ash to the organic substance in the insoluble portion

<sup>1</sup> [Pigmentkalk in the German; it is the compound noticed in page 461.—G. E. D.]

<sup>2</sup> De Cholelithis diss. inaug. med. Lips. 1832.    <sup>3</sup> Journ. f. pr. Ch. Bd. 25, S. 89.

<sup>4</sup> Zeitschr. f. rat Med. Bd 4, S. 193-208.

of gall-stones is altogether variable; in the insoluble part of six different concretions, there were 8·5, 12·1, 16·6, 30·4, 46·3, 50·6, and even 54·7% of ash; in the analyses of these six ashes there was comparatively much carbonate and little phosphate of lime according to the smallness of the ash; that is to say, in proportion as organic substance preponderated in the insoluble residue of a concretion, so much the more was the phosphate of lime encroached upon by the carbonate. In the ash which amounted to 8·5, there were 7·994 parts of carbonate of lime, and only 0·492 of earthy phosphates; while in the ash which amounted to 54·7, there were only 12·135 parts of carbonate of lime, a portion of which originated from oxalate of lime, which was recognized in the fresh object. Bramson has pointed out that dilute acetic acid extracts lime from the insoluble residue of biliary concretions; as this lime cannot be combined with sulphuric or oxalic acid, and as only an extremely minute quantity can be associated with phosphoric acid, it must be obtained from a combination with an organic substance: and as there is too little mucus present for us to ascribe it to that cause, it must necessarily have existed in combination with the pigment.

Further, if the bile-pigment were not in combination with some substance, it would be soluble in alcohol; for it is by no means a modified pigment which has become insoluble through some molecular change, but actual cholepyrrhin, in combination however with lime; and if we remove the lime by the application of a dilute acid, we obtain the cholepyrrhin, which is then soluble in alcohol, and possesses all the properties which we formerly enumerated.

An enormous deal has been written on the *formation* of the different varieties of biliary calculi, as well as regarding the proximate cause of the deposition of solid particles, and especially of the cholesterin; but any analyses of the various hypotheses that have been brought forward in relation to these points, would be here altogether out of place. The following is all that is actually known regarding the mode of formation of the concretions. Mucus and epithelium generally yield the points or foci around which a deposition of solid particles occurs; we always find pigment-lime with a little mucus in the centre of the concretion, and hence we may fairly conclude that it plays a part in their formation; but the separation of cholesterin from the bile is still not explained, even though mucus and pigment-lime can and must act as solid points. The question suggests itself, whether the bile lying amongst the gall-stones is normal in its character: it has been believed that it presents nothing abnormal,<sup>1</sup> but no conclusions can be drawn from any analyses of human bile that have yet been instituted, for the quantity of bile obtained from a dead body is too small to admit of an accurate analysis; moreover, the constitution of the bile when obtained after death, is generally more dependent on the morbid process which gave rise to the fatal termination, than on that which led to the formation of biliary concretions. It is, however, more than probable that in order that concretions of cholesterin should be formed, the bile should contain a smaller amount of the solvent for this substance than the normal fluid contains; but, as has been already mentioned, we very rarely meet with bile in

<sup>1</sup> Novi comment. acad. scient. inst. Bononiens. T. 3, p. 307-317.

which there is a separation of minute tablets of cholesterin, although they often occur in other fluids, as, for instance, dropsical effusions, &c.; hence the presence of solid insoluble particles must be regarded as exercising a considerable influence on the formation of gall-stones. If we inquire what it is which holds the cholesterin and the pigment-lime in solution in normal bile, direct experiments afford an answer to the question, and show that both these substances are principally held in solution by taurocholic acid or taurocholate of soda. If we digest the insoluble residue of a brown gall-stone with taurocholic acid or acid taurocholate of soda, it is entirely dissolved with the exception of a few grayish-white flocculi, and the previously colorless solution assumes the tint of fresh bile. Strecker showed long ago that cholesterin was soluble in solutions of taurocholic acid and its salts. Glycocholic and cholic (Strecker's cholalic) acids possess this property in a far less degree. The question regarding the formation of gall-stones would be very readily answered if it could be proved that bile which has a tendency to form concretions, was either poor in taurocholic acid in relation to cholesterin and pigment-lime, or that its taurocholic acid was decomposed in the gall-bladder, and had thus lost its power of dissolving these two substances.

Since concretions, which are rich in cholesterin, are never entirely devoid of pigment-lime, while, on the other hand, calculi which are poor in cholesterin, are always very rich in pigment-lime, the idea suggests itself that this latter compound takes an active part in the primary formation of these concretions; indeed, the frequency of their occurrence in certain districts in which the water abounds in calcareous salts, and in old age (when, as is well known, there is an increased tendency to all kinds of calcareous deposits, and when the separation of cholesterin is promoted by the attenuation of the animal juices,) seems strongly to favor this view.

We at present possess very few results, upon which the slightest reliance can be placed, regarding the *quantity of the biliary secretion*. By proceeding on perfectly different assumptions, some physiologists have calculated the amount of bile secreted in the human subject in twenty-four hours, at only one ounce, while others have considered that it amounts to as much as twenty-four. Blondlot, from his observations on dogs, in which he had established fistulous openings into the gall-bladder, calculated that one of these animals secreted between 40 and 50 grammes of bile in twenty-four hours; and hence that the amount secreted by man during the same time, would be about 200 grammes [or between 6 and 7 ounces].

Bidder and Schmidt<sup>1</sup> have obtained the following results regarding the absolute quantity of the bile secreted in 24 hours by the animals on which they experimented. For one kilogramme's weight of the animal there were secreted.

	In cats.	In dogs.	In sheep.	In rabbits.	In geese.	In crows.
Of fresh bile, . . . .	14.500	19.990	25.416	13.684	11.784	72.096
Of solid constituents in it,	0.816	0.988	1.844	2.47	0.816	5.256

<sup>1</sup> [The experiments of Bidder and Schmidt, which are here briefly referred to, are given in considerable detail in the new edition.—G. E. D.]



Nasse,<sup>1</sup> who made very numerous observations upon a single dog, obtained rather a higher mean number for the amount of bile than Bidder and Schmidt, who made numerous experiments on different dogs; there being, according to Nasse, 21·025 grammes of fresh bile, containing 0·746 of a gramme of solid constituents, secreted in 24 hours for each kilogramme's weight of the animal.

Experiments made on dogs led to precisely the same results as those upon cats [mentioned in p. 474]; the secretion reaching its maximum between the thirteenth and a half and the fifteenth and a half hour after the last meal. Greater fluctuations were, however, observed in the gradual augmentation of the biliary secretion in dogs than in cats.

The circumstance that the quantity of secreted bile, after attaining its maximum in the fifteenth hour after the last meal sinks with extraordinary rapidity, and even below the number which expresses the biliary secretion in the first hour after taking food, was confirmed by Bidder and Schmidt in their still more numerous experiments on dogs.

The same observers have likewise convinced themselves that when animals remain for a longer period than 24 hours without food (48, 72, 168, or 240 hours), the biliary secretion continuously diminishes, the daily diminution being, however, gradually less in proportion to the time that has elapsed since food was last taken. Thus, for instance, in cats, after 10 days' fasting, the biliary secretion amounted to only the fourth part of the quantity yielded in the 24 hours succeeding the last meal.

It was repeatedly observed by Bidder and Schmidt, and the observations have been confirmed by Nasse, that animals with permanent biliary fistulæ generally have a ravenous appetite. This circumstance may assist us in determining the question, whether the biliary secretion bears a definite proportion to the quantity of food that is taken. The question has been decided in the affirmative by the experiments of the first-named inquirers, and a series of observations by Nasse also confirm this view. Thus, for instance, Bidder and Schmidt found that when cats were overfed, the quantity of bile that was secreted exceeded by one-fifth the quantity which is commonly secreted by a cat after a moderately abundant meal. In these cases the augmented secretion of bile was, moreover, accompanied by an augmentation of its solid constituents.

From the preceding observations it might be expected that the *nature of the food* would exert a certain influence on the amount of the hepatic secretion; and this expectation has been thoroughly confirmed by the experiments of Bidder and Schmidt, and of Nasse. A *flesh-diet* induces a far more abundant secretion of bile than vegetable, amylaceous food. Thus, for instance, Nasse's dog, when fed on bread and potatoes, daily secreted 171·8 grammes of bile, containing 6·252 grammes of solid matter; but when fed upon flesh it secreted in the same period 208·5 grammes of bile, containing 7·06 grammes of solid matter. In admirable coincidence with these experiments are those instituted by Bidder and Schmidt on cats, which, when fed on pure *fat*, secreted no more bile than if they had been completely deprived of food for the same time. An *exclusive fatty diet*, therefore, exerts no influence on the secretion of

<sup>1</sup> Commentatio de bilis quotidie a cane secreta copia et indole. Progr. Marburgense, 1851.

bile. In Nasse's case, however, an abundant addition of fat to the ordinary food of the dog occasioned a marked augmentation of the biliary secretion.

In repeated experiments both on cats and dogs, Bidder and Schmidt found that, after the copious ingestion of water, the quantity both of the bile and of its solid constituents was increased. After water has been freely taken the bile is certainly somewhat richer in water than normal bile, but with this water there is at the same time secreted a larger amount of solid constituents than is usually eliminated by the liver. This result has also been confirmed by Nasse. Hence it is not surprising that slight variations are perpetually being observed in the ratio of the water to the solid constituents of the bile secreted in definite times; and hence, too, it is that in the numerous tables drawn up by Bidder and Schmidt, all influences on the hepatic secretion are far more distinctly and precisely reflected on the amount of the solid constituents than on that of the fresh aqueous bile. Nasse lays special stress upon the point, that the variations which we observe in the quantity of the solid constituents of the bile are chiefly induced by the organic matters, while the mineral substances secreted in definite times remain nearly constant.

After large doses of *carbonate of soda*, Nasse observed a considerable diminution of the secretion of bile, and especially of the solid constituents. *Alcohol* caused an augmentation of the fluid bile, but a diminution of its solid constituents.

Finally, Nasse entirely coincides with Bidder and Schmidt, that *disease* (namely, febrile excitement) has an extraordinary effect in diminishing the quantity of the secreted bile.

We must not overlook this opportunity of noticing the observations which Bidder and Schmidt have made regarding the *intermittent emptying of the gall-bladder*. Magendie first made the observation, that, after prolonged fasting, the gall-bladder is distended with very concentrated bile; Bidder and Schmidt have now convinced themselves that the gall-bladder does not empty itself immediately after the ingestion of food, but  $2\frac{1}{2}$  or 3 hours later; the mere distension of the stomach cannot therefore occasion the discharge of the contents of the gall-bladder. It must not, however, be inferred from this circumstance that all animals possessing a gall-bladder only effuse bile into the intestine during the period of digestion, and that at other times all the secreted bile is accumulated in the gall-bladder. For far more bile is secreted during the intervals between the individual meals than could be held in the gall-bladder; thus, for instance, the gall-bladder of a full-grown cat cannot contain more than about 3 grammes of bile, although the animal secreted in 24 hours from 30 to 32 grammes of bile, and therefore far more than could be collected in the gall-bladder, even with four or five emptyings after the ingestion of food. And the fact is still more strikingly shown in rabbits; the gall-bladder of a rabbit weighing 1 kilogramme can contain at most 0.469 of a gramme of bile; but since this animal sends 7 grammes of bile into the intestine in one hour, it is hence still less possible to conceive that all the bile must take its course through the gall-bladder.

Bidder and Schmidt have investigated this subject in a most accurate and ingenious manner. They arrive at the conclusion, from a large number of experiments on cats, that a cat weighing one kilogramme [nearly three pounds], when its digestion is most perfect, that is to say, when its biliary secretion is most abundant, secretes 0.765 of a gramme of fluid bile, corresponding to 0.050 of a gramme of solid residue in an hour; while, after ten days' fasting, there is secreted in the same interval only 0.094 of a gramme of fluid bile, yielding, when dried at 100°, a solid residue of 0.0076 of a gramme.

The secretion of bile is continuous; but, as is shown in the above cases, it is augmented or diminished according to the state of the digestion. Bidder and Schmidt found that the secretion attained its maximum ten or twelve hours after a copious meal, and from then till twenty-four hours after the meal, it gradually diminished, till it attained the same quantity which was secreted one or two hours after eating. In prolonged starvation, the quantity of the secreted bile gradually and progressively diminishes.

Thus, for instance, if a cat, weighing one kilogramme, discharges 0.492 of a gramme of fresh bile (obtained direct from the common biliary duct through an introduced canula) during the second hour after feeding, the quantity increases so rapidly, that during the fourth hour it secretes 0.629, during the sixth 0.750, the eighth 0.825, and in the tenth hour, 0.850 of a gramme of bile; so that, from the second up to the end of the tenth hour, the quantity of bile secreted increases, on an average, by 0.045 of a gramme in an hour. Moreover, the diminution in the secretion of bile takes place somewhat rapidly after the end of the tenth hour, the average hourly decrease from this maximum to the end of the twenty-fourth hour being 0.028 of a gramme.

With the view of determining the quantitative relation of the biliary secretion to the other animal excretions—a point of the greatest importance in estimating the physiological value of the bile—Bidder and Schmidt have instituted a series of statistico-analytical experiments on dogs, on about forty cats, thirteen geese, and several sheep and rabbits, in which biliary fistulæ had been instituted. They first determined the amount of carbonic acid expired by these animals, and then ascertained the ratio in which the secreted bile stood to it; and the result of these laborious investigations is, that “only from 1-10th to 1-40th of the carbon separated by the lung is secreted in an equal time by the liver in the form of bile, so that at least 8-9ths or 9-10ths of the burned and expired combustible materials do *not* pass through the intermediate stage of bile, but remain in the circulating blood, where they become thoroughly oxidized.”

In order to be enabled to form a definite opinion regarding that much-disputed question, the physiological importance of the bile, it will be expedient previously to establish a view regarding the *origin* or formation of the bile, from the facts from which science has as yet supplied us. The biliary secretion has always been regarded either as a pure function of the digestive process, or as a definite factor in the general economy of the animal organism. The difficulty of deciding between these views seems half removed when we have a clear understanding regarding the

formation of the bile, that is to say, regarding the substances from which its proximate constituents are formed. We have already seen that unfortunately there is still considerable obscurity regarding the origin of the individual substances which constitute the bile. Nevertheless, we trust to find in certain positive experiments and observations, a logical justification for either one or the other hypothesis. That which applies to the individual constituents, applies also to the bile collectively; the following facts will, however, probably indicate the path by which we may arrive at a knowledge of the mode of origin of the bile. The first point in this investigation is, to decide the question where the formation of the biliary constituents actually takes place, that is to say, whether they exist ready formed in the blood, or whether they are first formed in the secreting organ. The larger number of well-confirmed facts tend to show that the principal constituents of the bile are primarily formed in the liver itself from certain constituents of the blood conveyed to this organ by the portal vein. On comparing the histological formation of the liver with that of the kidneys, we perceive that in the liver there cannot be a pure transudation—a mere process of filtration of certain constituents of the blood—such as occurs in the kidneys. We know that in the liver the most minute blood-vessels are separated from the smallest canals which convey the bile by a thick layer of tolerably large cells, and that consequently in every case the substances given off from the blood must pass through cells endowed with vital force, before they can enter into the biliary canals. No comparison can be instituted between these cells and the epithelial cells which occupy the ducts of Bellini; for these hepatic cells close the extremities of the biliary canals (whether these form blind and distended sacs or very minute loops); if the smallest biliary canals possess a *membrana propria*, these cells, united in rows and having a coecal arrangement, lie external to it, and consequently in this respect differ essentially from the epithelial cells of the *canaliculi contorti* of the kidney, which take no part whatever in the urinary secretion. But the microscope which reveals to us the contents of these cells, indicates that they are elaborated from materials resorbed from the blood; for in addition to the round nucleus occurring in these cells, they contain a greater or less quantity of small molecules and vesicles, which very often become developed into distinct fat-globules; in numerous cases, however, these hepatic cells are filled with a yellowish matter, which sometimes appears in the form of distinct and separate molecular granules, and sometimes as diffused masses. With regard to the colorless fat-globules, they must necessarily undergo a metamorphosis within the cells, since very little free fat is generally found in the bile. From certain microscopical observations which I made in reference to the morphological contents of the hepatic cells of dogs and rabbits at different periods after taking food, it seems to follow that their physical characters vary with the stage of the digestive process. These and other histological relations which have been observed by Meckel<sup>1</sup> and Leidy,<sup>2</sup> show that it is in these cells that the substances taken up from the blood are elaborated into bile; and most of the physiological facts with which we are at present acquainted, accord with

<sup>1</sup> Müller's Arch. 1846.<sup>2</sup> American Journal of Medical Science, Jan. 1848.

this view. Müller, and subsequently to him, Kunde,<sup>1</sup> after separating the skin of the abdomen and tying the portal vein, opened the abdominal cavity of large frogs; ligatures were applied to all the points of attachment of the liver, and that organ was completely extirpated; after the operation, the animals were kept in narrow, dry vessels, at a low temperature, and the blood of those that were still surviving after two or three days, was collected by amputating their thighs. As we are justified in concluding, both from the experiments of Blondlot and from pathological observations, that icterus ensues within two or three days after the occlusion of the gall-ducts, we must here expect to find a very large quantity of bile-pigment and cholic acid, if the formation of the most essential biliary constituents take place externally to the liver; but although the examination was conducted with the greatest care, we could not detect, with certainty, any trace of either of these substances in this blood.

Moleschott has recently instituted a series of carefully conducted experiments on frogs in relation to this point. Like Kunde, he extirpated the liver; but succeeded in keeping the animals alive for a longer period. He could not succeed in detecting a trace of the resinous acids or of the pigment of the bile either in the blood or in the lymph, or in the flesh, or in the urine of the frogs on which he operated. It may, therefore, be regarded as an established fact, that the essential constituents of the bile are primarily formed within the liver.

I must here remark, that at the commencement of these experiments we believed that, though we could find no bile pigment, we had detected biliary acids; but we subsequently convinced ourselves that frog's fat, and indeed any fat that abounds in olein, yields with sugar and sulphuric acid a reaction extremely similar to that of cholic acid. But after we had become acquainted with this source of error, and had, as far as possible, removed the fat, no trace of bile could be recognized either by Pettenkofer's test or by any other means (as, for instance, the exhibition of taurine, the determination of sulphur in the alcoholic extract, &c.)

It certainly cannot be denied that after such severe operations, conclusions should only be drawn with the most extreme caution; but when taken in association with the above-named histological and with the physiological and pathological facts presently to be mentioned, the result to which we have been led by our experiments is deserving of a certain amount of weight.

It is further known that the biliary secretion differs from all other secretions in this respect, that it proceeds from the capillary system of a vein, and that even the blood of the hepatic arterial branches has become venous before it comes in contact with the finest ramifications of the biliary ducts; for, as Kiernan was the first to show, the *vasa vasorum* of the hepatic artery enter into a venous plexus which, instead of opening into the hepatic veins, discharges itself into the smaller (but not the smallest) branches of the portal vein, and in this manner forms the hepatic origin of the portal system. Hence the secretion of the materials of the bile takes place solely from pure venous blood. The secretion in the kidney, for instance, is altogether different, to which arte-

<sup>1</sup> Diss. inaug. Berol. 1850.

rial blood, and with it the substances (such as urea, uric and hippuric acids, &c.), which are first rendered excrementitious by the respired oxygen, are carried, and where, without having to pass through a dense layer of cells, these substances are transmitted in a manner very similar to simple transudation from the bloodvessels into the urinary canals. From the extreme slowness with which the blood passes through the liver, it follows that the conversion of the constituents of the blood into bile in the hepatic lobules only takes place very gradually, thus allowing of a more thorough and complete metamorphosis. (At these lobules, we find that the finest capillary network of the portal vein is separated by the plexus of the hepatic cells from the smallest biliary canals, which, according to E. H. Weber, are far more minute than the finest capillary vessels.) If we consider that the blood of the portal vein has been already collected from a capillary network, and that now, without further mechanical assistance, it has again to overcome the resistance of friction in a second capillary system, and further, that the veins into which the portal branches discharge themselves, are even deficient in the valves which usually aid the circulation within the veins, we can comprehend why it is that the blood passes very slowly through the liver. Müller and E. H. Weber have convinced themselves of the correctness of this assumption by direct microscopic observations on frogs and on the larvæ of salamanders. With these facts in our possession, we need be as little surprised that Bidder and Schmidt perceived that two hours elapsed after the administration of food, before there was an augmentation of the biliary secretion, and that it was not till the end of ten hours that the *maximum* flow took place, as at the great frequency of hyperæmic affections of the liver and of the associated congestion of the hæmorrhoidal veins.

If, however, the great slowness of the circulation within the liver forces us to the assumption that there is a peculiar elaboration of the materials in question within the hepatic cells, so also does the source of the substances which are conveyed into the portal blood, point to the peculiar function of the liver as a metamorphosing organ. The venous blood proceeding from the stomach, the whole intestinal canal, and from the mesentery, is collected in the portal vein; hence a great part of the nutrient matters absorbed in large quantity by the veins of these parts is conveyed into the liver; moreover the veins of the pancreas, and (what is more essential) those of the spleen, pour their blood into the portal vein. We shall presently see, when treating of the chemical and physical investigations of the blood, that the character of the portal blood varies according as the portal vein receives most of its blood from the stomach and intestinal canal during the process of digestion, or from the splenic veins, which convey a fluid very different from other venous blood. We shall, however, also see that the blood of the hepatic veins is as different from portal blood (whether collected during fasting or while the digestive process is going on) as from the blood of any other portion of the venous system. The blood within the liver, in its transition from the arterial into the venous state, undergoes more striking alterations than in any other organ. These changes are not confined to the mere abstraction of individual constituents from the blood in the liver,

but as we shall presently see, some of its constituents have undergone very distinct chemical changes. To this we must add, that the presence of the most important constituents of the bile cannot be recognized as preformed in the portal blood, notwithstanding many assertions to the contrary: at all events I have never succeeded in detecting them, even when operating on very large quantities.

The principal arguments against the view that the bile is formed from heterogeneous constituents within the liver itself, are based partly on the supposed analogy between the biliary and the renal secretions, and partly on certain pathological phenomena. That the analogy between the renal and hepatic secretions is limited to the single fact that they both are secretions, is sufficiently obvious from what is known regarding the difference in the structure of the two organs; and in reference to the facts derived from pathology, these, upon the whole, rather accord with the view that the bile is formed in the hepatic cells than that its actual constituents pre-exist in the blood. Jaundice very seldom occurs in diseases of the parenchyma of the liver, and almost never in the different forms of fatty degeneration or in tuberculosis of the liver, and very rarely in simple and red atrophy, in granular liver, and hepatitis; while the only diseases in which it is almost constantly present are those of the biliary ducts and acute yellow atrophy. If an accumulation of actual (chemically recognizable) biliary matters were induced in the blood by the suppression of the hepatic secretion, jaundice would necessarily occur just as frequently in the above-named diseases, which affect the parenchyma of the liver, as an impeded excretion of the bile. It is true that these diseases rarely attack the whole parenchymatous structure (indeed, hepatitis never does so, and jaundice seldom occurs in this affection), so that a portion of the liver could always provide for the separation of the bile from the blood: but again, on the other side, the facts may be urged that, in association with jaundice, there may be an abundant flow of bile into the intestine (as, for instance, may occur in pyæmia, yellow fever, after the bites of poisonous snakes, and even in cases of pneumonia accompanied by icterus), and especially that jaundice may occur in diseases in which no organic change either of the parenchyma or the gall-ducts can be detected. At all events this much is obvious, that we are unable to draw any conclusions from the presence of jaundice regarding a disturbance of the secretion of the liver or the separation of bile, and that it yields us no means of arriving at an opinion regarding the suppression of the biliary secretion or the formation of bile in the liver. Positive data are still required in order to enable us to decide, from the occurrence of jaundice, whether there is a mere separation or a secretion of bile; the different conditions which accompany or give rise to its occurrence are still so little investigated, that we are by no means justified in concluding that there is a formation of true bile in the blood, even in such cases as acute yellow atrophy of the liver (in which, in addition to the sudden access of jaundice, we find even the hepatic cells atrophied and destroyed).

In connection with this subject we would merely direct attention to some few points which have hitherto not been sufficiently regarded, in reference to pathological conditions. Thus, for instance, it is still

undecided whether other biliary matters, and more particularly the coagulated resinous acids, are found in the blood simultaneously with icterus; and it would even appear probable, from certain observations, that icterus may be present when no biliary acids are found in the blood. If it could have been shown which biliary acid,—that is to say, whether a conjugated acid or cholic (Strecker's cholalic) acid or choloidic acid—occurred in the blood of persons affected with icterus or in healthy individuals, it might have been determined whether its resorption was effected from the liver through the lymphatics or from the intestinal canal; but owing to the uncertainty of Pottenkofer's bile-test, our conclusions regarding the presence of the biliary resinous acids must be wholly subjective. The lymphatics undoubtedly play a highly important part in the resorption of the bile, and these vessels are moreover alone able to absorb bile from the liver, as the venous plexuses of the hepatic artery open into the portal vein, and would therefore convey the recently absorbed bile back to the hepatic cells. In the dead body the bile readily infiltrates into the neighboring parts, but in living animals such is not the case; it is probable, however, that we might also observe a similar imbibition of bile during life if it were not directly absorbed by the lymphatics surrounding and intersecting the surface of the liver as well as the biliary ducts and the gall-bladder. It is believed that many substances undergo chemical changes in the lymphatics; but it is not known whether bile-pigment and the biliary acids are carried unchanged through the healthy lymphatic system, or whether they experience any alterations in it. We do not know, therefore, whether or not the lymphatics perform their function in those diseases in which icterus is present without any obvious organic changes in the liver, or where, in addition to the jaundice, a large quantity of bile passes into the intestine. It would appear from experiments of injecting filtered bile into the veins, that the blood possesses the property, when in a normal state, of exerting a metamorphic action on the biliary matters; yet life may be prolonged for years after the complete occlusion of the ductus choledochus. We are, however, ignorant whether the blood in febrile and inflammatory conditions—where its oxidation is considerably diminished—loses the capacity for metamorphosing these biliary matters like the extractive matters, uric acid, cystine, &c. Why does icterus occur in fatty liver only when acute diseases supervene? In granular liver many of the small biliary ducts may be obliterated, and the hepatic granules consequently filled with bile, and yet icterus may not have been manifested during life. Acute yellow atrophy of the liver is a disease that has been but seldom observed, and still less investigated (excepting by Rokitsansky); of the chemical metamorphoses by which it is attended we know nothing. We do not think, therefore, that the meagre observations hitherto made by the bedside and in the dead-house justify us in drawing conclusions regarding the formation of bile in the blood, and the occurrence of icterus from the suppression of the biliary secretion.

If the view that the *formation of bile* takes place in the liver itself appears to derive considerable probability from anatomical and physiological facts, and is certainly not refuted by our pathological observations, we are necessarily induced to compare the juices conveyed to the



liver with those flowing from it; since it is only by a comparison between the fluids entering and leaving the liver\* that we can hope to attain certain and fixed points of support for a chemical survey of the mode in which the bile is prepared from different organic elements, and thus avoid too wide a deviation from the truth. If it be admitted that the portal vein mainly supplies materials to the liver, we must seek in the blood of this vein for the substances which contribute towards the formation of bile; and when the advanced state of our chemical knowledge shall enable us to institute a comparison between the constitution of the blood of the portal vein and that of the hepatic veins, it will necessarily be the means of elucidating the mode of formation of the bile and the function of the liver.

Unfortunately, however, chemical analysis is not in a sufficiently advanced state to afford a satisfactory reply to all, or even to many of the questions which we hope to solve by its aid; but yet it affords us the means of confirming or refuting some of the arguments advanced in support of one or the other of the above views. As we propose, in our remarks on "the blood," to enter more explicitly into the consideration of the different parallel analyses which we have made of the blood of the portal vein and of the hepatic veins, we will limit ourselves in the present place to a mere notice of the results in question.

The comparison between these two kinds of blood is probably more disturbed by deficiencies in our chemico-analytical appliances, than either by the admixture of blood originating from the hepatic artery with the blood of the hepatic veins, or by the abstraction of materials by the lymphatics. As far as concerns this addition of the blood of the hepatic arterial branches when it becomes venous, this is very small; for, independently of the small calibre of the hepatic artery, which is far less than that of the portal vein (a section of the hepatic artery is 4,909 square lines, while that of the portal vein measures 38,484 square lines, according to Krause and Valentin), the rapidity of the circulation of the blood in the veins proceeding from the hepatic artery must be nearly as small as in the equally large capillaries of the portal vein. The lymphatics, however, appear chiefly to absorb the material resulting from the nutrition of the vessels and the biliary ducts by the hepatic artery, and to carry off some portion of the previously formed biliary substances. On these grounds, it is necessary that a stringent comparison should be instituted between the blood of the veins which enter and leave the liver.

In passing to the consideration of the individual biliary substances, and inquiring which of these exist preformed in the blood of the portal vein, we find that none of the most essential constituents of the bile can be detected in it. The presence of resinous biliary acids, and therefore chiefly of cholic or choloidic acid, in the portal vein, has been conjectured even by those who do not believe in the formation of these acids in other parts than the liver; nor was their presence here to be wondered at, since there appeared to be grounds for assuming that a portion of the bile effused into the intestinal canal was resorbed by the veins, and that the rudiments of this resorbed bile must then of necessity be again collected in the portal vein; yet the most carefully conducted inquiries, instituted under various conditions, have hitherto failed to demonstrate the exist-

ence of these resinous biliary acids in the blood of the portal vein. The error of supposing that biliary substances have been demonstrated in the blood of the portal vein by means of sugar and sulphuric acid, arises from the similar reaction which Pettenkofer's test gives with olein and oleic acid.

We endeavored in pp. 120 and 240, to demonstrate the chemical grounds on which we based our hypothesis that cholic acid must be regarded as a conjugated acid, consisting of a non-isolable modification of oleic acid and a carbo-hydrate. We were led to adopt this view mainly in consequence of the large quantity of olein contained in the blood of the portal vein, in which respect it differs so greatly from the blood of every other vein, including even the hepatic veins. A careful examination of the blood of the portal vein, and a comparison of this blood with that of the hepatic and other veins, lead almost involuntarily to the conclusion that the oleaginous fats which occur in preponderating quantity in the blood of the portal vein, and are only contained in very small quantity in the blood of the hepatic veins, must participate to a considerable extent in the formation of the bile; for the blood is rich in olein when it enters the liver, but exhibits only a very small portion when it leaves that organ; the fat of the blood of the hepatic veins is also more consistent, and contains relatively more margaric. On an average, the solid residue of the portal blood contains 3.225% of fat, while that of the hepatic venous blood contains only 1.885%.

I do not purpose reverting here to the chemical grounds which appear to support the view that cholic acid is formed from oleic acid, but would simply observe, in relation to this subject, that I have never succeeded in producing sebacic acid by dry distillation from cholic acid, and, on the other hand, that I have convinced myself that not only the acids of the butyric acid group (Redtenbacher), but likewise those of the succinic acid group, more especially lipoic and suberic acids, may be produced from cholic acid by the action of nitric acid, in the same manner as from oleic acid (Laurent). Moreover, Kunde, as already observed, found that not merely the fat of frogs, but also every other animal or vegetable fat which is rich in olein, yields an intense purple-violet color with sulphuric acid and sugar; it does not occur with fats that are free from olein, and is most perfectly exhibited with pure oleic acid. This reaction of the oleic acid only differs, according to my experience, from that of cholic acid in occurring more slowly, and requiring the access of atmospheric air. As I did not perceive that there was any absorption of gas, I thought that the oleic acid might be contained in the cholic acid, in the modification of elaidic acid; but the latter acid yielded the same reaction as cholic acid with sugar and sulphuric acid, although less rapidly.

M. S. Schultze<sup>1</sup> has recently made the same observation with regard to olein. He also showed that the protein-bodies yielded a similar violet color when treated with sugar and sulphuric acid; and I have noticed the same circumstance in many ethereal oils, as, for instance, oil of turpentine and oil of caraway.<sup>2</sup> Pettenkofer's test is, however, by no means

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 71, S. 270.

<sup>2</sup> Kunde, De hepatis ranarum extirpatione diss. inaug. med. Berol. 1850.

to be rejected on this account, as it merely requires the same amount of caution in its application that is indispensable for the exhibition of every other chemical reaction.

The fat of the hepatic venous blood, when treated with sugar and sulphuric acid, yields the same reaction as that of the portal blood; and, when the experiment is conducted with care, we can scarcely fail to arrive at the conviction that no bile is contained in either kind of blood. The reaction does not ensue excepting with that portion of these two kinds of blood which is soluble in ether, and fails with those extractive bodies which are only soluble in alcohol—a fact which plainly indicates that these substances are not of a biliary nature.

The positive experiments, made under different physiological relations, by Bidder and C. Schmidt, on the biliary secretions of animals, appear, at first sight, to refute this hypothesis. These careful observers found that fat animals yield considerably less bile than lean ones, and that, when they were fed on fat (bacon, suet), the quantity was smaller than in the case of animals fed on substances containing the smallest possible portion of fat. These differences were no longer appreciable in animals which had been kept for some time without food. The fact that fat animals yield less bile than lean ones, is in harmony with an observation already referred to, that the metamorphosis of matter is usually accomplished more slowly, and in a smaller degree, in organisms disposed to secrete fat in abundance; we need only mention that fat animals expire less carbonic acid in equal periods of time than lean but strong ones. The inference to be drawn from these observations is, not that fat animals and fat persons yield little bile because they are fat, but rather that such animals and persons have grown fat in consequence of their secreting little bile.

It can scarcely excite surprise that animals which are fed exclusively on fat should secrete less bile; for all fat is not applied to the formation of bile, neither is it fat alone which is employed for that purpose; for we shall presently see that fat constitutes only a *part* of the material necessary for the formation of bile. Daily experience, derived from the pathological observation of cases of fatty liver, shows us also that an excess of fat is prejudicial to the secretion of bile, for although the hepatic cells are often dilated to twice the normal size in these cases, the quantity of bile is very much below the normal standard.

Lastly, the circumstance that animals which are fasting secrete more bile than those which are fed exclusively on fat, does not refute this hypothesis; for, as *much* sugar arrests the progress of vinous fermentation, *much* fat also impedes the formation of bile in the hepatic cells. As far as we are able to observe the metamorphosis of tissue in animals which have been kept for a long time without food, it would seem that this change is not limited to those histological elements which contain nitrogen, for we find that the fat rapidly disappears during inanition. We have already spoken, in the earlier pages of this volume, of the possibility of the formation of fat from the protein-bodies. If the observations of the above-named distinguished experimentalists do not admit the interpretation we have endeavored to give to them, the circumstance that the quantity of fat entering the liver is greater than that which passes from it, must remain entirely unexplained.

Sugar, or, at all events, a carbo-hydrate, must be regarded as another element essential to the formation of bile. The chemical equation according to which cholic acid may be regarded as composed of oleic acid and sugar, would not possess a higher value than any other one that might very readily be established from the high atomic weight of cholic acid, were it not supported by other grounds. We mentioned p. 258, that Bernard and Barrès<sup>1</sup> had found sugar in the tissue of the liver; and this discovery, which I have verified by my own observations, has been recently corroborated by the numerous experiments of Frerichs<sup>2</sup> on the livers of animals and men. This observer convinced himself that the quantity of sugar in the liver was wholly independent of the nature of the food—so far, at least, that it was discovered in the liver of animals which had been fed for a long time on flesh alone. We considered, at p. 259 the grounds which render it probable that sugar may be formed in the animal organism from the protein-bodies. Scherer<sup>3</sup> has recently drawn attention to a peculiar kind of sugar, incapable of fermentation, and found in the muscular juice; and C. Schmidt<sup>3</sup> believes that a small quantity of sugar exists in all normal blood. More or less sugar is always conveyed by the portal vein to the liver during the digestion of vegetable food; for we know, on the one hand, that the sugar which is gradually produced from starch through the whole course of the intestinal canal, is principally absorbed by the veins; and, on the other hand, that the veins of the stomach, and of the small as well as the large intestines, are emptied into the portal vein; and we consequently find, on a careful examination of the blood of the portal vein of the larger herbivorous animals, that it generally contains some portion of sugar, whilst this substance, as far as my experience goes, is much less constantly to be detected in the chyle. We are, therefore, disposed to agree with Frerichs in assuming that the sugar found in the parenchyma of the liver contributes, together with other constituents of the portal blood, at least in part, towards the formation of bile, although a large portion of the sugar formed in the liver is carried through the hepatic veins into the general mass of the blood. If our hypothesis of the constitution of cholic acid be correct, we can scarcely be surprised that sugar should lose 6 atoms of water in conjugating with oleic acid, when we bear in mind our previous experiences regarding conjugated compounds; but, after this union, we can no more detect sugar, as such, in cholic acid, than glycine in hippuric acid. (Compare p. 175.)

C. Schmidt, although, as has been already mentioned, opposed to the view that bile is formed from fat, suggests the ingenious hypothesis, that in the metamorphosis of fat in the animal body, sugar and cholic (Strecker's cholalic) acid are formed from the neutral fats, that is to say, from the combinations of glycerine with fatty acids: it is certainly a fact of some interest that, when we assume that one-seventh of the hydrogen of the glycerine ( $C_3H_7O_3$ ), is replaced by 1 equiv. of oxygen, we obtain the formula for anhydrous grape-sugar ( $C_6H_6O_6$ ), and that when we take

<sup>1</sup> Op. cit. p. 881.

<sup>2</sup> Verhandl. der physik. med. Gesellschaft in Würzburg. 1850. S. 51–55.

<sup>3</sup> Charakteristik der epid. Cholera. S. 162.

the fatty acid,  $C_{48}H_{97}O_3$  (correspond to the general formula for the solid fatty acids,  $C_nH_{n-1}O_3$ ), and assume that 7 of its equivalents of hydrogen are replaced by oxygen, we obtain the formula for cholic acid,  $C_{48}H_{99}O_9$ . HIO.

In opposition to Bernard's "formation of sugar in the liver," C. Schmidt remarks that we find sugar in the blood of the vena cava, as well as in that of the portal vein; and in reference to this point, I must observe, that I have found far more sugar in the blood of the hepatic veins (as noticed in the chapter on "the blood"), than in that of the portal vein or the jugular veins. In five determinations of these varieties of blood from different horses, I always found from 10 to 16 times more sugar in the solid residue of the serum of the hepatic venous blood, than in the corresponding residue from the portal blood; indeed, when the animals had been kept for some time without food, I could find no sugar in the portal blood, while it could easily be detected in the hepatic venous blood, and its quantity could be determined by fermentation. There can, therefore, be no doubt that sugar is formed in the metamorphoses which the blood undergoes in the liver. Now, if we perceive an excess of fat and a deficiency of sugar enter the liver, and if we find that these substances emerge from that organ in an inverse proportion, it appears obvious and mathematically certain that, according to the above-mentioned hypothesis of Schmidt, the fat is decomposed in the liver into cholic acid and sugar. But, although the above facts correspond so well with this hypothesis, we must consider that a formation of sugar may likewise be due to other substances. We shall presently see that there are also nitrogenous substances which undergo decomposition in the liver, and we have already indicated the possible formation of sugar from such decompositions; and Scherer's discovery of inosite (muscle-sugar) renders this view still more probable. Finally, we at present know too little of the extractive matters, in which the portal blood is by no means poor, to feel justified in denying that some of them may be converted into sugar.

From all this it follows that, unless we would rest satisfied with mere chemical formulæ and equations, we are still very far from comprehending the individual stages of the metamorphosis of animal matter, and of recognizing the nature of the changes that ensue, and the formation of the different new substances: the number of observations is, however, daily increasing, which must confine the admissibility and number of hypotheses within narrower and narrower limits. Thus, from the facts at present in our possession, we cannot decide with certainty which of the hypotheses regarding the formation of cholic acid—whether Schmidt's, or the one I have propounded—approximates the nearer to the truth; probably neither presents a perfectly correct view of the actual case. The following circumstances seem to tell against Schmidt's hypothesis, and in favor of mine: unconjugated acids containing 9 atoms of oxygen, are, at all events, very rare in chemistry; oleic acid yields the ordinary reaction with Pettenkofer's test, which is not the case with the solid fatty acids (of the general formula  $C_nH_{n-1}O_3$ ); and (which is of most importance) there is far less oily fat (although relatively more solid fat) in the hepatic venous blood than in that of the portal vein. This

much only seems fully established from the experiments which have been described, that the liver is an organ in which sugar is formed.

We can very easily comprehend, and need hardly entertain a doubt, that the nitrogenous adjuncts of cholic acid (Strecker's cholalic acid) are formed from the regressive metamorphosis of the nitrogenous parts of the animal body, and therefore especially from the metamorphosis of tissue: but physiological chemistry should not merely indicate possibilities and probabilities in the animal processes, but it should, at all events for the future, teach us the chemical equations expressing the decompositions of the individual animal substances, and the manner and successive stages in which the metamorphoses occur. We are, certainly, still far from attaining this object, but it is time that we should endeavor to reach it by all the auxiliaries and forces at our disposal. Taking this view of the subject, it would be important to ascertain whether the adjunct which yields glycine or taurine, exists preformed (either free or in combination) in portal blood. In regard to the glycine, every attempt to detect it in portal blood has been unsuccessful, although as much as 450 grammes [about 15 ounces] was examined for this purpose; this experiment cannot, however, be definitely regarded as proving that no glycine occurs in portal blood, since the cause of the negative result may be dependent on the imperfection of our chemical analyses; but, at all events, the opposite view, in accordance with which the glycine of the cholic acid is first formed in the liver, is not overthrown by this experiment, and we shall presently point out the grounds which support the idea that the glycine is produced in the liver from the metamorphosis of nitrogenous matters. Moreover, we are equally unable to detect preformed taurine in portal blood.

According to F. C. Schmid,<sup>1</sup> the ash of portal blood is richer in sulphuric acid than that of blood from the jugular veins; we might be thus led to suppose that the sulphuric acid of the portal blood was applied in the liver to the formation of the sulphurous adjunct, but this is not the case. It is well known that the estimation of the sulphur in an ash-analysis is the most uncertain of any of the determinations in analytical chemistry, since it depends on various accessory circumstances (the mode of heating, the presence of carbon hard of combustion, or the absence of alkalis with which the sulphuric acid that is formed might combine), whether more or less sulphur is volatilized. In employing this inexact mode of determination, I was, however, unable to find the difference between the blood of the portal and the hepatic veins, which Schmid observed between that of the portal and jugular veins. The preformed sulphuric acid in the water-extract of the portal and hepatic venous blood appears to be variable; but as a general rule, I always obtained rather more sulphuric acid from the serum of hepatic venous blood than from that of portal blood: the augmented quantity in the first case is, however, only relative; for the serum of the portal vein, in becoming changed into that of the hepatic veins, not only loses much water, but also albumen, as we shall subsequently show. This much may, however, be regarded as certain, that the preformed sulphuric acid no more con-

<sup>1</sup> Heller's Arch. Bd. 4, S. 323.

tributes to the formation of the sulphurous adjunct, than it passes into the bile. (See p. 398.)

If, however, we compare the quantity of sulphur in the two kinds of blood by the application of the dry method of oxidation, as, for instance, by potash and nitre, we find that the residue of the portal blood is certainly the richer in sulphur. On an average, I found 0.393 of sulphur (all the sulphuric acid being calculated as arising from sulphur) in 100 parts of the solid residue of portal blood, and 0.331 of sulphur in 100 parts of that of hepatic venous blood. The sulphur which is applied to the formation of this adjunct is therefore as latent (unoxidized) or combined in the portal blood, as in the adjunct itself. It now remains for us to inquire—to what substance does it owe its origin?

In the spirituous extract of portal blood (after the residue has been already extracted with ether and alcohol) I found a substance which, on incineration with nitre, yields sulphur. (As it can also be obtained when the blood has been previously neutralized, it cannot depend on any albuminate of soda that may have been dissolved by the spirit.) Moreover, this sulphur-compound is also found in lesser quantity in the blood of the hepatic veins. It is possible that the taurine, which, as we know, is rich in sulphur, may be formed from this sulphurous extractive matter. The principal source of the sulphur of the bile, and especially of the taurine, might, however, be sought in the perfect disintegration of the fibrin in the liver. I shall show, when treating of “the blood,” that the quantity of fibrin in hepatic venous blood is almost imperceptibly small, and, indeed, that often I could discover no fibrin whatever in it. The substance which was calculated as fibrin by Schultz and Simon, in their analyses of the blood of the hepatic veins, could not have really been that substance, but must have been the cell-walls of the blood-corpuscles, deprived of their contents by water. Hence, whether or not the extractive matters contribute to the formation of the nitrogenous and sulphurous adjuncts of the cholic acid, it is by no means improbable that the fibrin of the portal blood is applied in that manner. But there is reason to believe that these adjuncts are primarily formed in the liver, not merely from their absence in portal blood, but also on the following purely chemical ground: we have seen that glycine and taurine are not to be regarded as existing preformed in glycocholic and taurocholic acids; it is, however, the ordinary rule (and only few exceptions are known to it), that the so-called conjugated compounds are not directly formed from the adjuncts into which they become separated on decomposition; chemical experience, therefore, renders it improbable that these conjugated acids should be formed from pre-existing taurine or glycine and cholic acid. Moreover, we can hardly expect that in the animal organism, where complex compounds are resolved into simpler ones (when the retrograde metamorphosis predominates), comparatively simple substances should unite to produce more complicated ones in the formation of excreted matters.

If we attribute to the fibrin which is decomposed in the liver a great share in the formation of the above-named adjuncts, we must also at the same time meet the objection which may be brought forward, that the fibrin may be applied to the formation of the young blood-corpuscles

which are found in such large numbers in the hepatic veins. I have certainly never found the characters of portal fibrin to differ so much from that of other venous blood, in newly-killed animals, as has been observed by F. C. Schmid;<sup>1</sup> it appears, however, to be less contractile and less dense than that of other venous blood. This, at all events, appears not to be the form in which it can be applied to the construction of tissues or blood-corpuscles. Moreover, we see from a comparison of the portal serum with that of the hepatic veins, that the albumen in the latter is considerably diminished, and has probably been employed in the formation of blood-cells. According to my investigations, the albumen is to the other solid substances in portal serum as 100 : 12·5, while in the serum of hepatic venous blood (where, moreover, the salts are diminished by about 0·3) the ratio is as 100 : 27·4. Moreover, hepatic venous blood contains, both absolutely and relatively, far less serum than portal blood; when, for instance, the intercellular fluid is to the moist blood-cells in the ratio of 100 to 150 (and this was the case when horses were killed five hours after feeding), the corresponding ratio in hepatic venous blood is as 100 : 330; or if (ten hours after feeding) the ratio in portal blood is as 100 : 35, the corresponding ratio in hepatic venous blood is as 100 : 138. Hence the portal blood, in its conversion into hepatic venous blood, loses a very considerable portion of its serum, while the latter parts with much of its albumen. The coagulable, soluble albumen of the portal blood has, therefore, in a great part passed over into the considerably augmented cruor which is formed by the blood of the hepatic veins. If it is not too bold an hypothesis to assume that this portion of the albumen is applied to the formation of the walls of the blood-corpuscles, this readily explains why the bile is so rich in sulphur: for, as we shall prove in a future page, the walls of the corpuscles of hepatic venous blood contain no sulphur.

Another important constituent of the bile is the *pigment*, which also cannot be detected preformed in the portal blood; and we have already shown (in p. 282) that in all probability, it is formed from the blood-pigment; we shall, therefore, not again revert to the grounds on which this possibility or probability rests, but will merely observe that, if cholepyrrhin be actually a product of the metamorphosis of hæmatin, the process, at all events in the normal state, takes place in the liver. It appears no mere image of the fancy, to regard the distorted, speckled, irregular blood-corpuscles in the portal blood of fasting animals, as cells that are growing old; for, at all events, we find that the blood-cells leaving the liver by the hepatic veins, exhibit precisely those characters which we ascribe to young blood-cells; hence the cells of the portal blood do not undergo rejuvenescence in the liver, but suffer disintegration in that gland, and their remains are in part (the iron, for instance) applied to the formation of new blood-corpuscles, and in part are converted into excreted matters; hence it is very conceivable, that the hæmatin loses its iron and becomes converted into cholepyrrhin, which is mixed in the biliary canals with the other constituents of the bile. In instituting several comparative analyses with both kinds of blood, I found in 600 grammes of portal blood-cells, 0·384 of a gramme of metallic

<sup>1</sup> Op. cit.



iron, and in the corresponding 760 grammes of blood-cells from the hepatic veins, 0.333 of a gramme of iron. Hence, however great may be the errors of observation, this much is certain, that the iron of the decaying blood-corpuscles of the portal vein is more than sufficient for the requirements of the young cells of the hepatic venous blood. If we regarded the view as tenable, that the quantity of iron in the blood-corpuscles, or in the hæmatin, has any influence on the color, we would here notice the difference of tint presented by the blood of the portal and of the hepatic veins. F. C. Schmid has directed attention to the dark brown, sometimes velvet-like black color of the clot of portal blood; the corpuscles of the blood of the hepatic veins always appear of an intense purplish violet color, when seen in thin layers—a color which I have never observed in portal blood, nor to the same degree in any other venous blood. Whether this deficiency of iron in the blood of the hepatic veins be simply dependent on errors of observation, or whether the missing iron must be regarded as having passed into the bile, are points which I will not venture to decide, although I have made three experiments which coincided very well with one another. Since, however, iron has been so often found in the bile, the difference in the numerical results is probably dependent on the nature of the changes going on in the liver, and hence a part of the iron conveyed by the portal vein is carried off through the liver into the intestinal canal. Moreover, I was unable to find any iron in the serum of the blood of the portal vein, when free from red blood-cells.

Of the remaining organic constituents of the bile, the *cholesterin* is that which has received the most attention. It occurs, as we have already seen, in normal blood (see p. 247); and it is contained in that of the portal vein, although, in consequence of the preponderance of true fat, it can only be recognized and measured by the microscope with much difficulty. But independently of this, the frequent occurrence of cholesterin in morbid products (namely, in serous, encysted exudations, as, for instance, hydrocele) without any simultaneous affection of the liver, or without the simultaneous occurrence of other biliary constituents in the collective juices, sufficiently indicates that this substance is a product of the general metamorphosis of tissue, and that the liver is merely the organ by which, in the normal condition, this liquid is separated.

We have already seen why the occurrence of gall-stones rich in cholesterin, will not warrant the conclusion that there is an increased formation of this substance. The separation of cholesterin from the bile depends only on mechanical causes, and is independent of quantitative relations. Were we inclined to assume that there was a cholesterin-diathesis, we should at all events be astonished to observe, that when we found an exudation consisting almost entirely of a pulpy mass of pure crystals of cholesterin, gall-stones were never, or very rarely, simultaneously present.

It is unnecessary to offer any remarks regarding the origin of the *fats* and the fatty acids of the bile, since we have so often referred to the abundance with which fat occurs both in the blood of the portal vein and in the hepatic cells; the saponification of the free fats proceeds also in

other places; as, however, the fatty matters of the bile are for the most part saponified, while it is chiefly unsaponified fats which are found in the fat-cells, it would seem as if the fatty acids of the bile were first formed in the hepatic cells.

This circumstance appears to us to be opposed to the dehiscence of the hepatic cells assumed by certain authors; for even in fatty liver, or in certain physiological conditions in which the hepatic cells are filled with fat-globules, we neither find that the bile contains a large amount of unsaponified fat or any augmentation of the saponified fat, which must have been the case if the hepatic cells burst and discharged their contents.

In connection with the *mineral substances* of the bile, we shall first notice the *alkali* which it contains, and which is combined with the conjugated biliary acids, with fatty acids, and with pigment. Since both the water-extract and the spirit-extract of the portal blood yield alkaline carbonates on incineration, we can easily comprehend the source of these alkalis. Moreover, the albuminate of soda, in its transmission into the blood-cells, must also lose soda, which may contribute to the saponification of the fats and the formation of the biliary acid. In examining the ashes of the blood-serum of the hepatic and portal veins, I have found about as much, and, indeed, often rather more alkaline carbonates in the former than in the latter; but it must be considered that the blood of the hepatic veins contains little more than half as much intercellular fluid as the portal blood, and that consequently the whole of the blood of the hepatic veins contains far less alkali in combination with organic matters, than the whole of the portal blood. The same relation holds also with the alkaline carbonates, which I have found to exist preformed (by the method described in p. 393) in both kinds of blood: to determine them quantitatively was impossible; but it appeared to me (and to several eye-witnesses, the experiments being frequently repeated) as if the portal blood, when placed under the receiver of the air-pump, began to evolve air-bubbles in a less rarified atmosphere, and more abundantly, than the blood of the hepatic veins.

The quantity of the soluble *phosphates* in the bile is extremely small: like the earthy phosphates, they principally arise from the mucus of the gall-ducts. I have not found a constant difference between the amount of soluble phosphates in the blood entering into and flowing from the liver: the earthy phosphates would rather appear to pass from the blood into the bile; at all events, I invariably found more earthy phosphates in the clot of portal blood than in that yielded by the blood of the hepatic veins.

With regard to the *alkaline chlorides* which abound in the ashes of bile, the different quantities occurring in the two kinds of blood sufficiently explain their origin; in the serum of the portal blood, which has a comparatively low specific gravity, we find from 0.28 to 0.31% of chlorine, while in the denser serum of the blood from the hepatic veins only about 0.22% is found. On the other hand, the amount of chlorine in the blood-cells is much the same in both kinds of blood, and averages about 0.165%. Hence a portion of the alkaline chlorides must pass from the serum of the portal blood into the growing or young cells of the blood of the hepatic veins.

The singular result at which Bensch and Strecker have arrived, namely, that the bile of the herbivorous mammalia contains almost only soda-salts, while the food of these animals is rich in potash and deficient in soda, may probably be explained in the following manner: potash-salts are abundantly separated by other organs of the herbivora, as, for instance, by the kidneys; but in the liver no such separation takes place, because the potash conveyed into it with the portal blood is used for the formation of the blood-corpuscles (for, as C. Schmidt was the first to show, the blood-cells are especially rich in potash); we have, however, just seen that a great part of the alkaline chlorides passes into the cells of the blood in the hepatic veins.

Lastly, I must not omit to mention that the blood of the hepatic veins always contains considerably less water than that of the portal vein, and that even after abundant drinking, the quantity of water in the blood of the hepatic veins is only very slightly augmented, while in the portal blood it is increased to an extraordinary degree. Hence it follows that this excess of water in the portal blood is effused in the liver into the biliary canals, and that the density of the secreted bile must be liable to extreme variation from the external physiological causes.

In horses that had not drank much for five hours after feeding, their were from 70 to 110 parts more of water in the portal blood than in the blood of the hepatic veins, the standard of comparison being 100 parts of solid residue. However, in the latter case, the blood of the hepatic veins was the more aqueous of the two.

It is possible that this mode of explaining the origin of the individual biliary constituents may be set aside by further experiments, but notwithstanding its obvious imperfections, we have ventured to bring it forward, seeing that the principal object of an hypothesis is, in our opinion, to stimulate other inquirers to fresh investigations.

The following may be regarded as a brief abstract of the above views regarding the origin of the bile: while the non-nitrogenous and nitrogenous matters conveyed by the portal vein—most of which, even when in the blood, bear the character of substances in the process of metamorphosis—are applied to the formation of the biliary constituents, substances also pass into the bile, which must be regarded as the residue or secondary products of the process which gives rise to the formation or rejuvenescence of blood-cells in the liver; in the latter class we must especially place the fats and certain of the mineral constituents, while the nitrogenous substances, fibrin and hæmatin, are the most important members of the former. Hence we do not regard the bile as the product of the metamorphosis of any single morphological or chemical constituent of the animal body (neither of the fat-cells nor of the albuminates); but we believe that several substances, chemically and morphologically distinct from one another, undergo alterations in the liver, and that their individual products unite in the nascent state, and thus form the compounds and admixture of substances which we find in the bile.

In order that we may not omit any element which may contribute to our knowledge of the functions of the bile, we must still consider what finally becomes of this secretion in the intestinal canal; as, however, this

subject will be discussed in the chapter on "the intestinal juice," it will suffice here if we merely communicate the result of our experiments. The bile becomes gradually decomposed in the course of the intestinal canal, the conjugated acids breaking up and forming choloidic (or cholic) acid, which become converted into dyslysin—a substance that may be traced even into the rectum and the fæces; although the amount of biliary residue of this kind diminishes in the lower portion of the intestinal canal to such a degree that we are almost compelled to adopt Liebig's view, according to which the resinous constituents of the bile are for the most part resorbed from the intestine into the vascular system. Notwithstanding that the intestinal veins opening into the portal system, and the lacteals, afford the only means by which these biliary matters might again enter the blood, I have never succeeded in detecting the presence of such substances either in the chyle or (as has been already mentioned) in the portal blood in the normal state during the process of digestion. Hence, if there is no fallacy regarding the small quantity of dyslysin found in the solid excrements, we should be compelled to assume that the already modified biliary matters, absorbed by the lymphatics, were so changed in the glands that they no longer admitted of detection by the chemical means at present at our command.

The bile-pigments, although very much modified, are also found in the solid excrements, in addition to cholesterin and taurine. The soluble mineral constituents of the bile return from the intestine into the mass of the juices, as was long ago shown by Liebig.

After the above facts, and the conclusions based upon them, we need only say a few words in order to arrive at a judgment regarding the numerous, and often opposite, views in reference to the *functions of the bile*. We will, however, first briefly notice the opinions which we have already laid down regarding the physiological value of this secretion. Formerly, the point chiefly contested was, regarding the *excrementitious* or *non-excrementitious* nature of the biliary secretion, while all agreed pretty well in considering that the function of the liver was to purify the blood by separating the bile from it. We have endeavored to show, in a former part of this work (see p. 37), that a separation of zoochemical substances into secretions and excretions is both inexpedient and illogical, and it is therefore unnecessary to enter into any further discussion on this point. The view that the secretion of the bile is for the purpose of purifying the blood, needs no special refutation, since it is devoid of any logical justification; for such metaphorical indications of imaginary processes, and such vague analogies, are expunged from the physiological inquiries of the present day. For the benefit of those, however, who are unable to give up the old view, and who still regard the bile merely as an effete carbonaceous matter which the respiration does not remove, it may be mentioned that the bile—a secretion also not poor in nitrogen and hydrogen—is not separated in any increased quantity when the process of oxidation in the lungs happens to be disturbed; that there are no pathologico-anatomical facts which favor the view that the liver can act vicariously for the lungs; and, lastly, that the separation of carbon by the liver, as compared with that by the lungs, is so trifling (as is shown by the researches of Bidder and

Schmidt, noticed in p. 474), that the liver can hardly be regarded as essentially a blood-purifying organ, in so far as the elimination of carbon is concerned.

With regard to the *importance of the bile in the process of digestion*, and especially in chylication, it need hardly be observed that very different views have been advanced respecting the manner in which the bile acts upon the substances passing from the stomach into the duodenum. The oldest view is that which was advocated by Boerhaave, and originated by Sylvius de la Boe, according to which the bile contributes its alkali to saturate the acids of the chyme; and it does not appear to us to be open to so many objections as we usually find brought against it. It is certainly quite true that the bile can directly contribute little or nothing to the neutralization of the free acid; on the one hand, because the smallest quantity of acid added to the bile at once renders it acid; and on the other, because we find the chyme in the intestine still acid after the bile has been well mixed with it. The following appears to be what actually takes place: the alkali of the bile, occurring in combination with the resinous and fatty acids, must unite with the stronger acids of the chyme—namely, hydrochloric, lactic, and butyric acid,—and the resinous biliary acids which are thus liberated communicate an acid reaction to the chyme (as may be perceived by the application of litmus paper), until they are decomposed into dyslysin, or the insoluble resinous acids deprived of their adjuncts. Hence, in one point of view, the bile certainly contributes to the neutralization of the free acids contained in the chyme. This subject will be more fully considered in the chapter on “the intestinal contents.”

There is likewise another view regarding the uses of the bile in the intestine, which hardly deserves to be totally rejected. Haller was the first who ascribed to the bile the property of *dissolving fat*; the bile, however, only possesses this property in a slight degree, although one of its constituents, the taurocholate of soda, certainly has this power, as has been shown by Strecker. The bile, in consequence of its viscosity, undoubtedly promotes the disintegration of the fat into minute molecules; but, even in this respect, it is exceeded by other fluids. Hence we might believe with Frerichs, that the bile, at all events in association with the pancreatic juice, contributes to the perfect disintegration of the fat, and thus considerably promotes its absorption; and the results of several of the earlier experimenters, who, after tying the ductus chole-dochus, found an almost limpid, instead of a milky (fatty) chyle, would support this view. On the other hand, Bidder and Schmidt, in their experiments (which will be subsequently described), could observe no difference in the injection of the lacteals and the opacity of the chyle of animals to whom fat had been given, whether the bile was allowed to enter, or whether it was excluded from the intestine.

Some writers (and especially Hünefeld<sup>1</sup>) have ascribed to the bile a great *power of dissolving the chyme*, but neither starch, nor coagulated protein-bodies, nor any other of the constituents of the chyme, are essentially changed, even when digested for a long time with fresh bile; indeed, as a general rule, no change seems to be effected in these sub-

<sup>1</sup> *Chemie u. Medecin.* S. 105.

stances till the putrefactive process is set up by the biliary mucus. On the other hand, the water effused with the bile must not be disregarded as a solvent for the soluble portions of the chyme; we have already seen that the blood of the hepatic veins is always much poorer in water than that of the portal vein, and that the latter fluid often contains an extraordinary quantity of water; this water must necessarily often circulate from the intestinal veins into the portal vein, and from it, through the biliary ducts, back into the intestine, and must thus the more contribute to the gradual separation and extraction of the chyme, in consequence of its again losing in the intestine the substances it had taken up from the liver, owing to the insolubility of the biliary acids. This water is therefore differently freighted, according as it flows from the liver to the intestine, or from the intestine to the liver; or, so to speak, it percolates two different filters, each of which is only permeable for certain substances.

Moreover, it has been attempted to show that the bile exerts a general chemical action on the contents of the intestine, but these inquiries have led to directly opposite views. Some have considered that the bile exerts an *antiseptic* action on those constituents of the intestinal canal which have a tendency to decomposition; while others, again, ascribe to the bile the property of imparting a *definite direction to the metamorphosis* of these substances by its own decomposition. To those who would rest satisfied with such general views of the subject, we may observe, that the first of these opinions is, at all events, untenable; pure bile may certainly exert an antiseptic action on substances which become readily decomposed, as flesh, &c.; the bile, however, which is effused into the intestinal canal is not pure, but is mixed with mucus, which very readily undergoes decomposition, and, in point of fact, is actually decomposed in the intestine, as may be seen by the simplest observation. Hence we might, perhaps, be supposed to concur in the second view, according to which a definite character is impressed upon the metamorphosis of the food by the bile as a special ferment. But it must be frankly confessed that the assumption of *fermentative actions* in any process is nothing more than an indication of our positive ignorance. We shall, therefore, now proceed to the more special investigation of the metamorphoses which the individual constituents of the chyme undergo in consequence of the action of the bile.

We must by no means overlook the circumstance that the intestinal contents, when no bile is mixed with them, very soon undergo putrefactive decomposition; at all events, it has been found (by Frerichs) that, after tying the ductus choledochus, the contents of the intestines of animals thus operated on became completely putrid; and the same thing has sometimes been observed in patients with jaundice. In these cases Frerichs found in the bowels the substance yielding a rose-red color with nitric acid, which was discovered by Bopp amongst the putrefactive products of albuminous bodies. No great weight, should, however, be attached to this circumstance, in so far as the digestive process is concerned, since (as has been shown in the experiments of Schwann and Blondlot) animals in whom the ductus choledochus was tied, and whose bile escaped externally by an artificial fistula, lived and discharged normal excrements for months.

The view brought forward by H. Meckel, that bile converts sugar into fat, has been refuted by several experimenters, and is no longer supported even by Meckel himself.

He digested bile with sugar, and after the digestion he found more ether-extract in the bile than in bile not digested with sugar. The cause of the error may be easily perceived: ether-extract is not fat; the metamorphosis of the bile with its mucus is promoted by sugar, for the non-nitrogenous resinous acids (which are not insoluble in ether) are then formed more rapidly and in larger quantity than when sugar has not been added.

Prout is of opinion that the digested protein-bodies are converted by the bile into coagulable albumen, and Scherer<sup>1</sup> has made an ingenious experiment by which he thinks he has confirmed this view; lastly, Frerichs<sup>2</sup> has repeatedly seen filtered chyle become coagulable on the addition of bile and the application of heat. These experiments, although not the slightest doubt can be cast upon their accuracy, cannot be quite convincing, since it is, at all events, objectively difficult to prove that, on the one hand, the whole of the albumen which already existed, and was only prevented from coagulating, was previously removed from the chyme, and that, on the other hand, the turbidity of the mixed fluid was not dependent on the decompositions and reactions of individual substances, but on true coagulation of albumen by heat or of casein by acetic acid. I treated the purest peptones of albumen, fibrin, and casein which I could prepare, with bile and other reagents, but failed in obtaining a substance coagulable by heat or acetic acid, although I modified the experiment in numerous ways. Frerichs himself attaches no great value to this reproduction of albumen by the bile, for he remarks that only the smaller part of the ingesta dissolved by the gastric juice finds its way into the intestinal canal; by far the greater quantity passing directly from the stomach into the blood, and consequently being not at all exposed to the action of the bile.

Scherer introduced a mixture of bile and of flesh which had been dissolved in gastric juice into a portion of washed small intestine, and after tying both its ends, suspended it for some time in distilled water at a high temperature; he then found coagulable albumen in the water surrounding the intestine. As Valentin suggests, it is possible that in this a little albumen might be extracted from the vessels and glandules of the gut, even though it had been well washed with water.

After so many fruitless attempts to establish on incontestable grounds the co-operation of the bile in the *digestion of fats*, Bidder and Schmidt<sup>3</sup> have at length succeeded in submitting the question to the most exact proof. We shall follow these investigators through the different steps of the experimental proof by which they established the point. Experiments on dogs, in which they formed fistulous openings into the gall-bladder after having previously tied the ductus choledochus, showed that the bile which is poured into the intestine is devoid of any influence on the digestion of albuminous matter and of starch. Animals which had been thus operated on digested the same quantities of albuminous food

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 40, S. 9.

<sup>3</sup> Verdauungssäfte und Stoffwechsel. S. 215-234.

<sup>2</sup> Op. cit. p. 836.

as sound animals in which the bile could run unimpeded into the intestine, and in each case the process seemed to be equally perfectly performed. Precisely the same was observed in regard to amylaceous food; but the case was very different when the quantities of fat were compared with one another which were retained in the body and applied to the purposes of life by the animals that had been operated on and by the healthy animals. It was ascertained by Boussingault (sec p. 229), and the fact has been confirmed by Bidder and Schmidt,<sup>2</sup> that the animal organism is only able to absorb a definite, and indeed a somewhat small quantity of fat from the intestinal canal. Several experiments on cuts have shown that the full-grown animal is at most able to take up 0.6 of a gramme of fatty food for every kilogramme of its weight during the 24 hours, while young animals absorb as much as 0.9 of a gramme. Similar experiments with a dog (which weighed 5 kilogrammes) showed that this animal had resorbed 446.9 grammes of fat in a week; consequently, every kilogramme's weight of the animal would be able to digest at least 0.465 of a gramme of fat in one hour, when plentifully supplied with that substance. These animals, however, absorbed much less fat when the passage of bile was entirely excluded from the intestine; in three series of experiments on these animals it was found that in one case, where the access of bile was prevented, for every kilogramme of the animal's weight only 0.093 of a gramme of fat was absorbed, in another case 0.065 of a gramme, and in the third case 0.21 of a gramme. It is very clearly seen from these experiments, that a certain quantity of fat will be absorbed independently of the presence of the bile, although this is  $2\frac{1}{2}$  times less in the most favorable cases than the amount of fat which is absorbed in conjunction with the secretion of bile. The opposite experiment of Blondlot,<sup>1</sup> in which he could scarcely detect a trace of fat in the excrements of a dog, having a biliary fistula, and which had been fed on very fat food, has been, for various reasons, and perhaps correctly, referred by Bidder and Schmidt to the fact that a free passage through the ductus choledochus may probably have been re-established in the animal. The participation of the bile in the digestion of fat must, therefore, be considered as settled beyond a doubt, although it cannot be wholly denied that a small portion of the fat may be resorbed independently of the co-operation of the bile.

As it is well known that the white color of the chyle is mainly owing to the amount of fat which it contains, the color of the chyle contained in the lacteals was observed after the bile had been excluded from the intestine; but this experiment was attended by different results. Brodie,<sup>2</sup> as well as Tiedemann and Gmelin,<sup>3</sup> thought that they had convinced themselves that after tying the common bile-duct, the lacteals contained a colorless, transparent fluid, notwithstanding the use of fat food, whilst Magendie,<sup>4</sup> and more recently, even Lenz,<sup>5</sup> in connection with Bidder and Schmidt, have seen the chyle appear milk-white under similar relations. If it may be *à priori* anticipated that we cannot form a

<sup>1</sup> Essai sur les fonctions du foie et de ses annexes. Paris, 1846, p. 52.

<sup>2</sup> Quarterly Journal of the Sciences and Arts. 1858, Jan.

<sup>3</sup> Die Verdauung nach Versuchen. Bd. 2, S. 24-48.

<sup>4</sup> Précis élémentaire de Physiologie. T. 2, p. 117.

<sup>5</sup> Op. cit. p. 58.



very definite opinion of the more or less white color, or of the greater or less transparency of the chyle contained in the lacteals, the uncertainty of this mode of observation must be doubly manifest to all those who have frequently observed the lacteals in animals which have been killed immediately after feeding. Hence it follows that, as we have already seen, even when the bile is excluded, a portion of the fat is resorbed, and renders the chyle more or less whitish. The quantitative determination was here the only way of deciding the question with certainty, and this was, therefore, the course which Schmidt pursued. In the chyle obtained from the thoracic duct of dogs with biliary fistulæ, he found on one occasion 0.834% of fatty acids mixed with other organic substances, and on another occasion 0.190% of free fat, together with 0.113% of fatty acids, while the chyle of a healthy dog, that 8 hours before its death had been fed upon beef, contained 3.244% of free fat, with 0.058% of fatty acids. While the differences in the amount of fat in these two kinds of chyle are so great, the other constituents were found to fluctuate very slightly in their quantitative relations. Moreover, this experiment perfectly confirms the fact which had been otherwise established, that the bile essentially contributes to the absorption of fat.

If it be rendered tolerably evident by these experiments, that the bile is indispensable to the absorption of fat into the juices of the animal organism, its mode of action in this process still remains unexplained: and this result must appear the more striking, seeing that direct experiments instituted with fat and bile afford no clue to the explanation of the mode of action. The bile possesses in a far less degree than the pancreatic juice the power of forming an emulsion, and even if it did possess this property in a well-marked degree, the resorbability would be by no means explained by the extreme comminution of the fat; for since the coats and cells of the intestine are continuously permeated with aqueous moisture, and can never be dry at any point, we cannot understand, from a physical point of view, how the oily fat can penetrate these membranes. Hence it has been assumed that the fat is saponified by the alkali of the bile; but since the greater part of the chyle-fat is unsaponified fat, we are compelled either to withdraw altogether from this hypothesis, or to assume with Moleschott,<sup>1</sup> that the fat is saponified in the intestine (by means of the pancreatic fluid), but is again liberated in the lymphatics. This latter view, independently of its teleological improbability, can hardly be accepted when we consider that after the use of fatty food, mere traces of fatty acids are found in the intestinal canal, that unsaponified fat is recognizable even in the epithelium and cells of the villi, and that, according to Schmidt's experiments, the exclusion of the bile renders the chyle very deficient in free fat, while it does not affect its quantity of fatty acids. Lastly, the bile possesses so very slight a solvent power (none whatever, according to Bidder and Schmidt) for neutral fats (and even for the fatty acids, it would appear from the experiments of Lenz, not to be very considerable), that the bile which is secreted would be perfectly insufficient to dissolve the whole of the fat which is resorbed. It has been consequently supposed that individual parts of the inner intestinal surface may be specially capable of absorb-

<sup>1</sup> *Physiologie des Stoffwechsels*. Erlangen, 1851, S. 203.

ing fat, and that fat alone can penetrate through them; but in that case the assistance of the bile in the resorption of the fat would appear to be altogether superfluous. But since the bile has been shown to be necessary to this object, nothing in fact remains but to assume that the bile induces a modification in the relations of adhesion between the oleaginous fluid and the moist watery membranes, by which the transmission of the fat through these membranes is effected. The theory of the physical relations of the different kinds of fluids to different membranes has been as yet so little studied, that such an assumption as the above is by no means inadmissible; indeed we find that Bidder and Schmidt performed an experiment which indicates with tolerable distinctness the existence of such a relation; they plunged two glass capillary tubes in oil, having previously moistened the interior of one of them with bile; the oil rose far higher in the tube moistened with bile than in the other, either when it was perfectly dry or when moistened with a saline solution. This mode of explaining the absorption of fat has been established beyond all doubt by the accurate experiments of Wistingshausen<sup>1</sup> (conducted under Schmidt's superintendence), 'on the relations of the fats when mixed with the acids of the bile to endosmosis and capillary attraction.'

Moreover the experiments made on animals in which fistulous openings were established between the gall-bladder and the external abdominal walls (by which means all the bile that was secreted escaped externally), which have led Schwann,<sup>2</sup> Blondlot,<sup>3</sup> H. Nasse,<sup>4</sup> and Bidder and Schmidt<sup>5</sup> to very opposite views, do not prove that the bile exerts any *very* great influence on the digestive process. If animals can live for two or three months, or even half a year, without the passage of bile into the intestinal canal, the function of this fluid in digestion must at all events be a very limited and probably only an indirect one, and this is the conclusion we should draw from the accurate and ingeniously devised experiments of Bidder and Schmidt; for, as has been already mentioned, the secretion of bile does not attain its maximum till the tenth hour after food has been taken, and by this time by far the greatest part of the ingesta has passed along the duodenum; hence the bile enters the small intestine at much too late a period to exert in it any great influence on the metamorphosis of the chyme. The biliary secretion unquestionably stands in a definite relation to digestion; a relation, however, which must be considered rather in the light of an effect or consequence of the digestive process than as an intermediate link in the process itself.

We are thus led back to the view to which we have often alluded, according to which the most important function of the liver is the *formation*, or at all events the *rejuvenescence of the blood-corpuscles*, a view which, as is well known, was long ago rendered more than probable by E. H. Weber, and more recently by Kölliker, by numerous histological investigations of foetal livers and of foetal blood, as well as of the livers of tadpoles. Although we shall enter more minutely in another place, when treating of "the blood," into the consideration of the different views that have been promulgated regarding the origin of the blood-cor-

<sup>1</sup> Dissert. inaug. Dorp. Livon. 1851.

<sup>2</sup> Müller's Arch. 1844. S. 127.

<sup>3</sup> Essai sur les fonctions du foie et de ses annexes. Paris, 1846.

<sup>4</sup> Handwörterbuch der Physiologie. Bd. 3, S. 837. <sup>5</sup> In a private communication.

puscles and the different situations in which they may be formed, yet as the subjects are so closely allied, it may be expedient here briefly to mention the grounds in favor of the above view. Since the consideration that the liver is a permanent factory for blood-cells appears, even to us, to be somewhat paradoxical for many physiological reasons, we shall allow the facts which present themselves as the immediate results of our comparative analyses of the blood of the portal and the hepatic veins, to speak simply for themselves.

The blood of the hepatic veins contains a far larger number of *colorless blood-cells* (the so-called lymph-corpuscles) than the blood of the portal vein. They do, however, also occur in the latter, although very scattered, in twos or threes at different points, and they are nearly of equal size, very coarsely granular, and on the addition of acetic acid exhibit a bipartite or tripartite nucleus. In the blood of the hepatic veins they present a very different relation: on an average calculation, their number is at least fivefold that of the colorless cells in the portal vein; they present extreme differences in size, varying from 1-306th to 1-180th of a line; they have for the most part a very faintly defined outline, are slightly granular, and often resemble colorless yolk-cells; the smaller ones are usually somewhat more clearly defined, and exhibit dots on their surface; the larger ones become much swollen in water, but at a certain degree of attenuation appear to collapse; they then form dark and very prominent granular masses under the capsules of the colored blood-corpuscles; the larger colorless cells swell very much on the addition of acetic acid, and then exhibit a *single*, large, lenticular, excentric nucleus. Moreover, these colorless cells, which are of the most varied sizes, are aggregated in groups of five, six, or seven.

As we must return to the fuller consideration of the colorless cells, when we treat of "the blood," and shall then, moreover, explain the reasons in support of the view that the red blood-cells are formed from the colorless ones, it will be sufficient simply to mention my own observations regarding these cells, in order to support, from this point of view, the claims of the liver as a blood-forming organ.

The *red cells* of the blood of the hepatic veins are altogether different from those of the portal blood; in regard to their grouping, I very often found them assume the nummular arrangement in portal blood (in horses five hours after they had taken food), while in the blood of the hepatic veins I could never find a trace of any such arrangement, but saw them lying together in irregular heaps; we have become acquainted, through the admirable investigations of F. C. Schmid, with the peculiar *color*, the spotted appearance, and the irregular *forms* of the colored cells of portal blood; nothing of the kind was ever discovered in the corresponding blood-cells of the hepatic veins; they presented a sharp outline, and a very slight, although a recognizable, central depression.

The *capsules* of the colored cells in the two kinds of blood present well-marked chemical differences, especially in their relation to water. The colored cells of ordinary blood, if watched under the microscope, almost entirely disappear when much water is added; this is also the case in portal blood, although here also, as in every other kind of blood, a few of the colored blood-cells, or rather of their capsules, still remain

visible. In the blood of the hepatic veins a very different state of things is, however, observed; on diluting it with from 30 to 50 times its volume of water, the blood-corpuscles certainly are changed, that is to say, they become pale, swell up, lose their pigment, and unite so as to form membranes which, under the microscope, resemble detached serpent's scales. We have previously observed, that these decolorized blood-cells in the blood of the hepatic veins, were formerly mistaken for fibrin; but we may readily convince ourselves by the microscope of the almost entire absence of fibrin in the cruor of hepatic venous blood, and by the following experiment, of the accuracy of this view, and of the great number of these indestructible corpuscles in the blood of the hepatic veins. On mixing the fluid expressed from the clot with 20 times its quantity of water, portal blood, like that from any other vein, yields a slight flocculent deposit, in which shreds of conglomerated cell-walls may be recognized by the microscope; if, on the other hand, we treat an equal volume of fluid strained from the cruor of hepatic venous blood with 20 times its quantity of water, there will be a flocculent precipitate of 6 or 8 times the bulk of the precipitate in the other experiment (although the non-fibrinous cruor of the hepatic venous blood contains in its interstices one-half more serum than an equal volume of any other blood); in this manner I obtained, after the most careful washing and boiling with alcohol, 0·245% of these cell-membranes from the clot of the portal blood, while from the hepatic venous blood, similarly treated, I obtained from 1·98 to 2·43%. This cell-membrane was perfectly insoluble in a solution of nitrate of potash (even after 48 hours' digestion at 35°); I was unable to discover sulphur in it by boiling it in potash-lye, &c.

If this behavior of the membranes of the colored cells of the hepatic venous blood indicates that there is here an excess of newly-formed or rejuvenescent blood-corpuscles, the proof that a formation of new blood-cells takes place in the liver is fully confirmed by a comparison of the *contents* of the corpuscles of the two kinds of blood. We find far less hæmatin in the cells of hepatic venous blood, than in those of portal blood; for on an average, 180 grammes of the moist cells of hepatic venous blood contain scarcely so much iron as 100 grammes of the cells of portal blood. On the other hand, there is more globulin or coagulable matter generally, and more chloride of potassium, but considerably less fat, in the cells of the hepatic venous blood, than in those of the portal blood.

In the blood of the hepatic veins, the *fibrin* is either entirely absent, or is present in mere traces; while in the portal blood, taken at the same time, we often find a perfectly normal, strongly contracting fibrin.

The *serum* is relatively much less abundant in the blood of the hepatic veins than in that of the portal vein; while in the latter there are 70 parts of serum to 100 of corpuscles, in the former there are only 32 parts of serum to a corresponding quantity of cells; if the portal blood happen to be rich in water, as, for instance, when there are 287 parts of serum to 100 of cells, the blood of the hepatic veins will, even then, not contain more than 73 parts of serum to 100 of cells.

The serum of the hepatic veins is certainly more concentrated than that of the portal vein; if we accurately compare the two, we find in the

former, a relative and absolute *diminution of the albumen* (there being in 1000 parts of the serum of the hepatic venous blood fully a third less albumen than in an equal quantity of the serum of portal blood), while on the other hand, as has been already mentioned, the globulin in the blood-cells is relatively and absolutely increased. The *phosphates, chlorides* and *potash-salts* are diminished in the serum, but are in excess in the cells of the hepatic venous blood. *Sugar* is relatively and absolutely more abundant in the serum of the hepatic venous blood, than in that of the portal blood.

If from these facts we should regard the liver as a seat of formation of blood-corpuscles, in which certain residua of this process are at the same time perfectly eliminated from the blood, and appear in the form of bile in the excretory ducts of this gland, the above-mentioned observations of Bidder and Schmidt would no longer excite our wonder. We can easily understand why the biliary secretion does not attain its height until ten hours after the ingestion of food, when we recollect the extreme slowness with which the blood circulates in the hepatic capillaries, and when we consider that the formation or rejuvenescence of the blood-cells would certainly require some time in order that they may attain the perfection they possess when leaving the liver by the hepatic veins, and that the secondary products (the biliary matters) naturally cannot be duly separated till this principal process is almost concluded. Hence it need no longer excite our wonder, that during foetal life the liver should possess so relatively large a volume, that the blood of the foetus is far richer in corpuscles than that of adults (Poggiale),<sup>1</sup> and that even during this period bile is poured into the duodenum, although there is nothing in the intestine to be digested. Assuming this view to be correct, we may further easily understand why, in hepatic affections, and especially when they arise in consequence of metallic poisons (which, as is well known, are most prone to localize themselves in the liver), the number of cells in the blood frequently appears to be considerably diminished.

If the bile is merely a secondary product of the formation of blood-cells in the liver, we need not wonder that the animals in Schwann's and Blondlot's experiments could live for so long a time without any considerable disturbance of the digestive organs or of the general health; and if these animals finally died when the bile was completely excluded from the intestine, their deaths might result from unobserved or unsuspected causes, such, for instance, as their licking up the bile, and thus disturbing the gastric digestion: but several of the above-mentioned circumstances show that the bile likewise discharges certain functions in the intestine, which without it are not so rapidly or so perfectly performed; thus, for instance, it promotes the finer disintegration of the fat contained in the food, hinders the chyme from undergoing putrid decomposition, purifies it, and saturates the stronger acids which have passed from the stomach into the intestine. Although any one of these influences, taken alone, would not be of sufficient importance to affect the vitality of the organism, yet when long continued and collectively, they may excite such disturbances in the animal economy as gradually to destroy life. Hence if the absence of bile in the intestine does not exert a direct dis-

<sup>1</sup> Compt. rend. T. 25, p. 198-201.

turbing influence on the vital processes, it may indirectly lead to the destruction of the organism, just as we see disturbances induced in the circulation from very trifling mechanical deficiencies of the valves, which indirectly, and perhaps after many years, give rise to fatal results.

Lastly, we have to mention a ground comparatively unimportant in itself, which is opposed to the excrementitious nature of the bile, and in part explains the injurious effects which gradually ensue when the secretion is altogether excluded from the intestine; we refer to the resorption of certain constituents of the bile—a circumstance which has been very prominently put forward by Liebig. It has been already mentioned that we are unable to detect any traces of resorbed bile either in the chyle or in the portal blood, but that a careful examination of the contents of the bowel, in various portions of the whole intestinal tract from above downwards, almost necessarily leads us to adopt the view that the greater part of the bile is again resorbed as it passes through the intestine, and is returned to the general mass of the fluids. If a circuit of the bile from the liver through the intestine, and from thence back into the liver, appear at first sight objectless or superfluous, teleological grounds should not restrain us from the recognition of positive facts, especially when we are still very far from being able to master the aims of nature or her method of arrangement. The chief biliary constituents which are resorbed, are the soluble salts and the cholic acid liberated from its adjunct. If, as we have previously endeavored to show, this acid is produced from fat and sugar, or solely from fat, it would be teleologically just as difficult to understand why this important element of nutrition or supporter of respiration, almost immediately after being taken, should again be given off to the external world. Whether the cholic acid be formed from fat or not, it does not at all possess the chemical constitution which true excrementitious substances usually present. In our general consideration of the animal substrata, we have already mentioned that there must always be a correspondence between the chemical and the physiological qualities of a body. Now, in its chemical qualities, and especially in the numerical relations of its atomic composition, cholic acid closely resembles, and indeed is perfectly identical with the true nutritious matters and respiratory elements; for sugar, dextrin, and lactic acid are far less complex substances, far more oxidized, and far poorer in carbon, than cholic acid, and yet no one entertains a doubt regarding their physiological value in reference to nutrition and the metamorphosis of matter. We cannot perceive why cholic acid should form so striking an exception to this rule. But if we have regard to the teleological objection, according to which it seems incongruous that this substance should first be removed from the blood in order again to be taken up into that fluid, we may reply to this that many useful and likewise useless substances are repeatedly separated from the blood by the salivary and gastric glands (as we see in the case of sugar, iodide of potassium, and the salts of ammonia), and are again taken up by it. In the repeated passage of iodide of potassium through the salivary glands, no one can doubt that the relations of transudation peculiar to those organs are responsible for this phenomena. We cannot, however, ascer-

tain what mechanical or chemical conditions in the liver, besides the formation of blood-cells, are necessary for the secretion of cholic acid in the minute biliary canals. The resorption of the cholic acid in the intestine should therefore not be deemed more unnatural or irrational than the resorption of the chloride of sodium which is separated with the bile. We are as unable to understand the special object which nature has in view in the resorption of the cholic acid, as we are to comprehend the metamorphoses which the resorbed bile appears very rapidly to undergo in the lymphatic vessels or in the blood. If it, therefore, only remains for us to assume that the resorbed cholic acid (as a respiratory element already partially consumed in the organism) contributes its part to the warming of the animal body, we have a further explanation why the perfect exclusion of bile from the intestine (in Schwann's experiments) may prove prejudicial, although very gradually, to the general health of an animal.

## THE PANCREATIC JUICE.

Notwithstanding the careful analyses of Tiedemann and Gmelin,<sup>1</sup> as well as those of Leuret and Lassaigue,<sup>2</sup> the pancreatic juice, until the last few years, has been one of the most imperfectly understood of all the fluids of the animal body; very recently, however, several excellent works have appeared on the chemical nature and the physiological function of this fluid. Bernard,<sup>3</sup> Frerichs,<sup>4</sup> and lastly Bidder and Schmidt,<sup>5</sup> have obtained from their investigations results which, although not entirely coincident, are so decisive and certain that the function of the pancreas is now more clearly understood than even that of the liver.

The pancreatic juice is a colorless, clear, very slightly tenacious fluid, devoid of taste and smell, having an alkaline reaction, and a specific gravity ranging from 1·008 to 1·009; on heating it, there is only an inconsiderable coagulum formed, and on the addition of acids and alcohol, it only becomes slightly turbid. The specific gravity of the pancreatic fluid is liable to considerable variations (Ludwig and Weinmann),<sup>6</sup> since the amount of its solid constituents varies inversely with the time during which the secretion has been going on; Frerichs, who examined a very dilute pancreatic juice, determined its specific gravity at 1·008 or 1·009, while Bidder and Schmidt found the specific gravity of a thick viscid specimen which they were investigating to be 1·0306.

In correspondence with this density of the pancreatic juice, Schmidt found that on one occasion it contained 9·92%, and on another 11·56% of solid constituents; in the former case there was 9·04 of organic matters, and 0·854 of ash, which contained 0·736 of chloride of sodium, the remainder being chiefly bibasic phosphate of soda.

<sup>1</sup> *Verdauung nach Versuchen*. Bd. 1, S. 28.

<sup>2</sup> *Recherches phys. et chim. pour servir à l'histoire de la digestion*. Paris, 1825, p. 104-108.

<sup>3</sup> *Arch. gén. de Méd.* 4 Sér. T. 19, p. 68-87.

<sup>5</sup> In a private communication.

<sup>4</sup> *Op. cit.* pp. 842-849.

<sup>6</sup> *Dissert. inaug.* Zurich, 1852.

This secretion is so prone to decomposition, that after exposure to the air for a few hours, it develops a distinct odor of putrefaction. Frerichs found 1.36% of solid constituents in the pancreatic juice of an ass, and 1.62% in that of a dog.

We have here quoted the properties of this fluid as they have been described by Frerichs, because we have ourselves obtained similar results in an experiment made on a large mastiff; moreover, the description given by Leuret and Lassaigue agrees pretty closely with the above. On the other hand, Bernard found this fluid very viscid and tenacious, and so rich in a coagulable substance that, on the application of heat, there was entire solidification—much the same as Tiedemann and Gmelin had formerly noticed; and, in correspondence with the above reaction, there were very considerable precipitates thrown down by alcohol, acids, and metallic salts. According to Bernard, the above-described very thin pancreatic fluid is only secreted when the gland has become inflamed in consequence of the injury attendant on the operation; but since, immediately after the operation, a very thin secretion is poured forth, poor in coagulable matter, Bernard's explanation cannot hold good, and the extreme fluidity of the juice can scarcely be referred to a diseased condition of the gland in question.

In order to obtain the pancreatic fluid, we must previously give a meal to the animal to be employed, and then make an incision two or three inches in length into the *linea alba*, for the purpose of seeking the duodenum after the abdominal cavity has been opened; the descending portion must then (according to Frerichs) be laid open, and the mouth of Wirsung's duct sought. If, as in the human subject, the bile-duct opens at the same spot as the pancreatic duct, the former, as a matter of precaution, should be tied, while a small silver canula should be introduced from the intestine into the latter, in order to obtain this fluid in a state of purity.

Bernard attempted to obtain the pancreatic juice by establishing a fistulous opening from Wirsung's duct; with this view, he cut through the duct near the point where it enters into the duodenum, and drew the cut end towards the abdominal walls, to which he attached it by sutures.

The principal constituent of the pancreatic juice is a *substance resembling albumen or casein*, but which is not perfectly identical with albuminate of soda, with casein, or with ptyalin. It coagulates only imperfectly when heated (probably from its containing an alkali), is precipitated by acetic acid, but redissolves slowly in an excess of the acid, and especially if heat be applied; it is precipitated from its acetic-acid solution by ferrocyanide of potassium; it is precipitated by nitric acid, and if it be then boiled, especially if ammonia has been added, it assumes a deep yellow color; on the addition of chlorine-water it separates in grayish flakes; it is thrown down by alcohol, but, according to Bernard, redissolves readily in water. Frerichs found 0.309% of this substance in the pancreatic juice of an ass. It is to this substance that the pancreatic fluid especially owes its principal chemical and physiological properties.

Bernard found a considerable quantity, and Frerichs a smaller amount (0.026%), of a *butter-like fat*.



The *organic matters soluble in alcohol* only amounted to 0·015% in the pancreatic juice of the ass.

Neither Frerichs nor Bernard could detect the presence of *sulphocyanides*.

In reference to the *mineral ingredients* (as determined by incineration), Frerichs found 1·01% in the secretion from the ass; of this amount, 0·12% was insoluble, and consisted of carbonate and phosphate of lime and magnesia, and 0·89% was soluble, consisting of chloride of sodium and alkaline phosphates and sulphates.

Nothing can be stated with any accuracy regarding the *quantitative relations of the secretion*, since the injury caused by the operation which is necessary for the purpose of observing the secretion must very much derange the physiological relations. The experiments of Frerichs merely show that it is only during digestion that the pancreatic juice is secreted. In a state of abstinence, Frerichs found the gland pale and anæmic, and the duct of Wirsung empty.

Frerichs collected 25 grammes from an ass in three-quarters of an hour, but only 3 grammes from a hound in twenty-five minutes, during the process of digestion; Bernard obtained 8 grammes from a large dog in one hour, and 16 grammes hourly after inflammation had been set up. Bernard found, as a general result, that in the latter case there was always an increased flow of the pancreatic juice, but that it ceased to be coagulable and viscid.

The quantity of the pancreatic fluid varies very much in different animals; according to Colin<sup>1</sup> it does not stand in a direct ratio to the volume of the gland. While the pancreas of the ox and of the horse yields 260 or 270 grammes in an hour, that of the swine, which is about half the size, yields only 12 or 15 grammes in an hour.

The recent observations of Bidder and Schmidt on the pancreatic juice of the dog differ considerably from those of Bernard [above referred to]. They found that a strong dog (weighing 20 kilogrammes) secreted 7·86 grammes in 8 hours and a quarter, there being 1·614 grammes secreted in the first hour, while in the eighth there was only 0·73 of a gramme. We must, however, observe that the secretion was only collected from the lower and larger duct, while the course of the fluid into the intestine through the upper and smaller duct was not impeded. From these observations on the dog, Bidder and Schmidt calculate that an adult man, weighing 64 kilogrammes [or about 10 stone], secretes 150 grammes in 24 hours.

Ludwig and Weinmann found in a series of experiments, which extended over 7 days, and included 37 observations, that a dog for every kilogramme's weight secreted 35·184 grammes of pancreatic fluid in 24 hours. The amount is, however, liable to considerable variations; prolonged hunger, vomiting, and operations on the animal diminish the amount, while the ingestion either of solids or fluids increases it. The quantity increases very rapidly after water has been taken; in two experiments the secretion attained its maximum in 12 or 13 minutes after drinking.

*Diseases* of the pancreas are, as is well known, extremely rare; I

<sup>1</sup> Compt. rend. T. 34, p. 85.

once found, in the duct of Wirsung, a concretion which exhibited all the characters of a protein-body, but differed from the better known salivary concretions in yielding very little carbonate and phosphate of lime, and indeed very little ash at all.

The *importance* of the pancreatic juice *in relation to digestion* was first recognized by Valentin; it converts into sugar the amylaceous matters which have not been metamorphosed by the saliva, and have passed unchanged into the duodenum. Valentin supported his opinion by the fact that the pancreas is much more developed in herbivorous than in carnivorous animals, and convinced himself that the expressed juice, or an infusion of the sliced gland, possesses in a high degree the power of converting starch into sugar. Bouchardat and Sandras<sup>1</sup> found that the juice discharged from Wirsung's duct by fowls or geese possessed this property, but lost it after being heated to 100°. They further ascertained that this property is peculiar to the nitrogenous or albuminous substance which is precipitable by alcohol, and afterwards soluble in water. More recently, this subject has been investigated with the greatest scientific accuracy by Bernard and Frerichs, to whose labors we have so often referred, as well as by Bidder and Schmidt; and it is now indubitably established that the pancreatic juice possesses this sugar-forming power in a far higher degree than the saliva.

Bidder and Schmidt have likewise shown that the matter of which the sugar-forming power of the pancreas depends, exists preformed in the fresh juice, and is not, therefore, formed as in the saliva, by the mixture of different fluids, and that it maintains its efficiency far below the temperature of the animal body, and does not even lose this power of metamorphosis when it has remained for 24 hours at a temperature of 18°, while its action on starch is not affected either by the bile, the gastric juice, or free acids.

In order to institute a comparison between the amount of force exerted on starch by the saliva and by the pancreatic juice, it would be absolutely necessary to make an accurate quantitative determination of the amount of starch, which may be metamorphosed by equal quantities of the two kinds of juices; but, unfortunately, determinations of this nature, however important they may be in other respects, have not been successfully accomplished. We believe, however, that we should no more over-estimate the metamorphosing action of the pancreatic juice than that of the saliva, for, although the action of the pancreatic juice may be somewhat stronger than that of the saliva, it is a striking fact, that we generally find many unchanged, or at most, merely transversely contracted starch-globules in the excrements of herbivorous, and even of ruminating animals (even when they have been sparingly fed upon amylaceous food for some days before they were killed). Since, on the other hand, Bidder and Schmidt have made the observation in the case of a sheep having a fistula in the abomasum, that only a small quantity of starch was found in its fourth stomach, we must necessarily regard the metastatic force of the pancreatic juice as somewhat limited. (I am bound to observe, that the presence of starch is always recorded in my

<sup>1</sup> Compt. rend. T. 20, p. 1085.

journal in reference to my various examinations of the contents of the stomachs of ruminating animals.)

The action of the pancreatic juice does not, moreover, appear to extend very far into the intestine. According to Bidder and Schmidt, it seems wholly to disappear in the upper half of the intestinal canal; at all events, the contents of the intestine are unable beyond that point to develop butyric acid from butter, which is a property of this juice.

Colin has already specially noticed the fact, that the amount of the secretion does not stand in a direct relation to the volume of the pancreas, and hence we should be cautious in drawing any conclusion as to the functions of this gland from the volume of the pancreas in different animals; besides, the volume of this gland is so different in different animals living on the same kind of food, that nothing, either for or against any view, can be deduced from the size of the pancreas: formerly it was generally assumed that the pancreas was on an average by far more voluminous in the herbivora than in the carnivora, but in the rabbit, for instance, the weight of this gland amounts to 1-600th part of the bodily weight. Bidder and Schmidt, who made the latter observation, assign to the carnivora the more voluminous pancreas, but this again is not strictly true, for while in cats, for instance, the weight of this gland amounts to 1-300th part of their bodily weight, in the beaver it amounts to 1-30th. (E. H. Weber.)

Bernard claims for the pancreatic juice another, and apparently, a more important function; he believes that he has found that it is solely by the action of the pancreatic juice that the fat is reduced to a condition in which it can be resorbed and digested; that is to say, that it is decomposed into glycerine and a fatty acid. This view, although it appears to be supported by convincing proofs, is, however, directly opposed by the numerous and ingeniously devised independent experiments of Frerichs, and of Bidder and Schmidt.

Bernard's experiments, which, strangely enough, were confirmed by the French Academy,<sup>1</sup> have reference to the following points. Both the excretory ducts of the pancreas were tied in dogs, and the animals were afterwards fed upon food abounding in fat; no milky chyle was found in the lacteals; the fat remained unchanged, and was found unaltered even in the large intestine. The following experiment appears even more decisive in favor of Bernard's opinion. If oil be injected into the stomach of a rabbit, which is afterwards allowed to partake of its ordinary food; and if it be killed three or four hours after the injection of the oil, Bernard maintains that none of the lacteals will be filled with milky chyle, except those which originate from the intestine below the opening of Wirsung's duct. This experiment would seem at the same time to show that the bile exerts no influence on the digestion of the fat, since in rabbits Wirsung's duct opens somewhat lower in the duodenum than the bile-duct. Finally, the pancreatic fluid, when shaken with fat, was said readily to form an emulsion in consequence of its viscosity, and to retain the fat in this finely comminuted state, far longer than any other animal fluid; the neutral fats, however, in a short time, being decomposed into glycerine and the corresponding fatty acids.

<sup>1</sup> Compt. rend. T. 28, p. 960.

It is singular that neither Frerichs nor Schmidt and Bidder, although their observations were made with the greatest care, have been able to confirm any one of these experiments, which Bernard maintains that he has often repeated. These experimenters have followed all Bernard's directions; after tying the pancreatic duct in cats, they have kept the animals without food for from twelve to twenty-four hours (so that it might be fairly presumed that there was no longer any pancreatic juice in the intestine), and then fed them only with milk, fatty food, or butter, killing them in from four to eight hours after the meal. These experiments were often repeated, and the lacteals were always most beautifully injected, and the receptaculum chyli distended with milky chyle.

Frerichs performed the following experiment on puppies and cats, which had fasted for a long time: he tied the small intestine far below the opening of the biliary and pancreatic ducts, and below the ligature he injected milk with olive oil, or an emulsion of oil and albumen, or pure olive oil, and he found, after two or three hours, that the lacteals were filled with white chyle. Frerichs, however, believes that he has found that the extreme comminution of the fat, and hence in some measure its resorption, are promoted by the bile and pancreatic juice; and he draws this conclusion from the following experiment:—in cats which had long fasted, he cut through the small intestine near the middle, injected olive-oil into both halves, and tied the two cut extremities; in this case, he found the lacteals springing from the upper part of the intestine always far more injected than those proceeding from the lower portion, and he referred this to the circumstance that the bile and the pancreatic juice had access to the oil in the upper part of the intestine; for, although pure pancreatic juice, when shaken with oil out of the body, reduces the particles of oil to a state of extreme minuteness, the latter soon separate again on the surface.

We have already remarked that Bernard's experiment is by no means convincing on the one hand, because the chyle contains far less fatty acids than the ordinary neutral fats, and on the other hand, because other animal fluids, as soon as they begin to putrefy, cause a similar decomposition of the neutral fats. Schmidt and Bidder<sup>1</sup> have, however, taken the trouble to prove in a direct manner the fallacy of Bernard's view by numerous experiments. After having fed cats with butter, they could find no trace of butyric acid in the contents of the intestine, in the chyle, in the blood, or in the bile. Hence, although decomposing pancreatic juice when in contact with butter, at a temperature of 37°, in the course of a few hours gives rise to the formation of butyric acid, no such formation of this acid occurs in the animal body. Schmidt and Bidder now tied the duodenum at its upper part, between the pylorus and the mouths of the pancreatic and biliary ducts, and by means of a pipette, injected melted butter immediately below the ligature, but above the mouths of the ducts; in the course of six or eight hours the contents of the intestinal canal certainly contained some butyric acid; the same occurred when the ductus choledochus was at the

<sup>1</sup> Quoted by Lenz (who co-operated with Schmidt and Bidder) in his Inaugural Thesis, *De adipis concoctione et absorptione*. Dorp. Liv. 1850.

same time tied. Hence the power which the pancreatic juice possesses in the formation of butyric acid, is impeded by the gastric juice. The converse experiment in the laboratory, with a specimen of pancreatic juice (from a large dog) at a temperature of  $37^{\circ}$ , shows that the gastric juice here acts only as a dilute acid, and may be replaced with a precisely similar result by equally diluted lactic, tartaric, or acetic acid.

Moreover, I have not found any confirmation of Bernard's experiment on rabbits (although it is one that may be easily repeated), in which he observed that after feeding them with fat, the milky injection of the lacteals could only be perceived beneath the opening of Wirsung's duct. Bidder and Schmidt have, however, discovered how Bernard was led into this error, for on injecting butter into the gullet of rabbits, they found that after two hours, the lacteals given off between the pylorus and the mouth of the pancreatic duct, were fully distended with milky chyle *very rich in fat*; if the animals were killed four hours after the injection of the fat, the lacteals situated eight or ten centimetres [about three or four inches] above the mouth of the duct were still filled; if they were killed six hours afterwards, only those below the mouth of the pancreatic duct were thus injected; and, finally, if they were not killed for eight or ten hours, the first lacteals well injected with milky chyle were found to be situated from 20 to 30 centimetres [from eight to twelve inches] below the opening of the duct. Hence it must have been by always killing the animals six or eight hours after feeding them with fat, that Bernard was able, apparently to maintain his view. The facts of the case were simply these. The chyle had already passed onwards from the lymphatics proceeding from the first portion of the duodenum, and there was no more fat to be absorbed in that portion of the intestine, when Bernard began the investigation.

Frerichs has also overthrown another and an earlier view of Bernard's, namely, that the pancreatic juice acidified with hydrochloric acid might take the place of the gastric juice in relation to the coagulated protein-bodies.

Lastly, Frerichs is of opinion that as the decomposition of the bile is very much hastened by the pancreatic juice, this property is of some importance in effecting the rapid conversion of the bile into insoluble products incapable of resorption.

## THE INTESTINAL JUICE.

Our knowledge is very slight regarding the fluids secreted by the glandular organs of the intestinal mucous membrane. This depends in a great measure on the difficulty of obtaining these secretions isolated from the remains of food, from the secretions of the liver and the pancreas, and from the products of digestion. On this point, also, Frerichs<sup>1</sup> has thrown some light; previously to his researches, our theories on this subject were based rather on subjective views than on objective facts.

<sup>1</sup> Op. cit.

Frerichs has shown that it is in the highest degree probable that the lenticular capsules which occur in the small intestine, partly as solitary glands and partly in heaps as Peyer's glands, only contribute slightly to the formation of the intestinal juice; these lenticular glandules are, as is well known, shut sacs, which only rarely, and for the most part when in a morbid condition, burst, and thus discharge their contents on the mucous membrane of the intestine. By exposing these minute sacs to the action of the compressorium, Frerichs ascertained that they contained an alkaline fluid, which was coagulated by acetic acid, the turbidity being dependent on the presence of molecular granules and morphological elements resembling cell-nuclei. In typhus and other conditions which are associated with intumescence of Peyer's patches, and with prominence of the individual spherical capsules, the correctness of these views may be very readily confirmed. Hence Frerichs is fully justified in regarding the pouch-like glands which in the small intestine are known as the glands of Lieberkühn, and in the colon, as the follicles of the large intestine, and which occur in great numbers, and are of very considerable size, as the true secreting organs of the intestinal juice. The chemical examination of the intestinal juice also shows that the fluids secreted in the small and in the large intestine are perfectly identical. Frerichs obtained this secretion for examination by applying ligatures to pieces of intestine from four to eight inches in length in cats and dogs, after having, as completely as possible, removed the contents of the gut by pressure; he then returned the intestine into the abdominal cavity, and killed the animal in four or six hours. In the piece of gut enclosed between the ligatures, there was then found a glassy, transparent, colorless, and tenacious mass with a strong alkaline reaction. In precisely the same manner I obtained the intestinal juice from the ileum of a man who, in consequence of a badly performed operation for hernia, had several intestinal fistulæ, with perfect inversion of a loop of gut; at one of these fistulous openings, fecal matter appeared; at the other, pure intestinal juice might be collected. The morphological elements found in the intestinal juice are granular cells in greater or less abundance, cell-nuclei, here and there a little fat, and not unfrequently cylindrical epithelium (in the case which I examined, the latter structure was very abundant). Notwithstanding this perfect coincidence between my experiments and those of Frerichs, which is the more striking since they were made in different ways, some recent investigations of Bidder and Schmidt<sup>1</sup> show that this subject obviously requires further elucidation; for by following the method indicated by Frerichs, these physiologists could obtain no trace of intestinal juice, so that they were compelled to postpone its examination till they could collect it from an artificially formed intestinal fistula in a dog, in whom the pancreatic juice and the bile were carried away externally by a corresponding fistula.

The gut, both in recently fed and fasting cats, was tied immediately below the duodenum, and exactly in accordance with the directions of Frerichs. Three or four loops, at the distance of half a foot, were isolated by ligatures, and replaced; the wound in the abdomen was then

<sup>1</sup> In a private communication.

stitched up, and in the course of from three to six hours, the animal was killed by strangulation. There was "not a drop" of intestinal juice to be found. In the dog from which they obtained the intestinal juice, two fistulous openings had been established, one from the gall-bladder, and the other from the small intestine to the external abdominal walls, which were perfectly healed in the course of ten days after the operation; by the introduction of silver and caoutchouc tubes, they obtained at the upper opening pure bile, and at the lower one the glandular secretion of the small intestine, mixed perhaps with a little unresorbed saliva and gastric juice, whose quantity, however, in the fasting state, would be so extremely minute as to be unworthy of notice.

The intestinal juice does not mix readily with water; it cakes, and apparently coagulates when treated with a saline solution, as an aqueous solution of chloride of sodium or sulphate of soda; the portion soluble in water behaves in precisely the same manner as the mucous juice, which will be described in a future part of this work. Frerichs found from 2.2 to 2.6% of solid constituents in the intestinal juice, in which the parts soluble in water amounted to 0.87%, the fat to 0.195%, and the ash to 0.84%; I found only 2.156% of solid constituents.

Frerichs has not succeeded in effecting a change in any of the ordinary elements of food by means of the intestinal juice. Protein-bodies and gelatigenous substances remained perfectly unchanged; fat became disintegrated just as in all other viscid fluids. Moreover, it exerted no special action on starch; at all events, after prolonged digestion at 37°, no more boiled starch was converted into sugar than would have been obtained by the action of animal membranes, soluble albumen, casein, &c. Hence Frerichs is compelled to deny to the intestinal juice any action as a direct digestive agent; but, on the other hand, the intestinal juice which I collected from the loop of gut of the patient in our hospital, possessed in a high degree the power of converting starch into sugar; but protein-bodies and fats, whether the juice were modified or not, were so little affected by this mucus, that I must express my doubts whether it exerts any digestive action on these substances; and the more so, since cubes of coagulated albumen and pieces of flesh, when introduced into the lowermost of the fistulous openings, were expelled from the rectum almost entirely unchanged; the fistula was, however, on the lower part of the ileum, and probably near the cæcum. Bidder and Schmidt have, on the other hand, convinced themselves, by the most striking experiments, that this *intestinal juice not only metamorphoses starch* with as great rapidity as saliva and pancreatic juice, but also that *the intestine exerts as powerful a digestive influence on flesh, albumen, and the other protein-bodies, as the stomach.*

In cats that had been kept for some time without food, the duodenum was cut below the openings of the pancreatic and biliary ducts and above a cork plug, which was inserted and strongly tied into the upper end, so that the secretions of the stomach, pancreas, and liver, were absolutely excluded; in the lower end two cylinders of flesh and albumen were sewed up in muslin bags, and pushed down as far as possible, and the wound stitched to prevent their escape; the gut was then replaced, the edges of the wound in the abdomen brought together, and in the

course of five or six hours the animal was killed. The muslin bags, which were found low down in the small intestine, appeared externally to be much collapsed; and, on opening them, the pieces of flesh and albumen presented a macerated appearance, as if they had been exposed to the action of a gastric juice, and were strongly alkaline; the albumen was thoroughly softened and broken down, and in the twelve experiments at present made, lost from one-eleventh to one-half of its original weight (the experiments including both albumen which had been dried at  $120^{\circ}$ , and moist specimens), so that in the latter case the contents of the bags appeared to have almost entirely vanished. The experiment succeeded equally well when the gastric juice was excluded, but access of the bile and pancreatic juice was allowed. The cork plug was then, of course, introduced between the pylorus and the openings of the biliary and pancreatic ducts.

Fresh, pure intestinal juice has hitherto been only examined by Bidder and Schmidt,<sup>1</sup> and (under their superintendence) by Zander:<sup>2</sup> it is a colorless, ropy, viscid fluid, which is invariably alkaline; the alkalinity, however, varies in different animals, and in different parts of the intestine; but no definite rule can be laid down on this subject.

The juice, after the removal, by filtration, of the morphological elements above mentioned contains no trace of albumen, and therefore, does not coagulate either on boiling or on the addition of acetic acid: alcohol of 85% throws down white flakes, which redissolve in pure water; their solution is precipitated by acetate of lead, but not by the mineral acids, or by bichloride of mercury: the acetate-of-lead precipitate dissolves readily in acetic acid.

According to Bidder and Schmidt, the filtered intestinal juice of dogs contains from 3.042% to 3.467% of solid substances.

Zander found 3.9% of solid constituents in a specimen of intestinal juice containing bile and pancreatic fluid; amongst the solid constituents there were 2.5 parts soluble in alcohol (glycocholate and taurocholate of soda), and 1.4 parts insoluble in alcohol (taurine, pancreatic fluid, and intestinal juice); the unfiltered juice contained 0.8% of epithelium, &c.

Bidder and Schmidt infer from the following observation, that the pure gastric juice must be a tolerably diluted fluid. The filtered intestinal contents, in which there are 3.8% of solid constituents, consist not only of the true intestinal juice, but also of gastric juice, bile, and pancreatic fluid; the gastric juice has about the same concentration as the fluid intestinal contents; but the bile of the dog contains 5%, and the pancreatic juice 10% of fixed substances; hence the intestinal contents could not attain to such a high degree of dilution, unless the true intestinal juice were an extremely aqueous fluid.

It is obviously impossible to form any certain determination regarding the *quantitative relation* of this secretion. Bidder and Schmidt calculated from the concentration of the mixed intestinal juice (that, namely, containing bile, gastric juice, and pancreatic fluid), and that of the gastric juice, the bile, and the pancreatic fluid, that the pure intestinal juice must contain about 15% of solid constituents, and that, consequently,

<sup>1</sup> Verdaunungssäfte und Stoffwechsel. S. 260-282.

<sup>2</sup> Diss. inaug. Dorp. Livon. 1850.



that an adult man (weighing 64 kilogrammes or 10 stone) secretes in 24 hours about 300 grammes of intestinal juice.

The quantity of the secretion naturally varies according to the period of digestion. In the dog, in which an intestinal fistula was formed in the middle of the small intestine, the following remarkable facts were observed by Bidder and Schmidt. This secretion flowed most abundantly from the fistula 5 or 6 hours after a meal; and its quantity was considerably increased very soon after drink had been taken: however, the most singular circumstance is, that the intestinal juice shows the same concentration as before the ingestion of the fluid; hence we must conclude with Schmidt, that the drink is absorbed in the stomach and in the upper part of the small intestine, and that the water, which thus finds its way into the blood, increases the intestinal juice in common with the other secretions.

With regard to the *functions* of the intestinal juice, it seems to a certain degree to unite in itself the powers of the gastric and pancreatic fluids. For it is established by the numerous experiments of Bidder and Schmidt, that this fluid can dissolve and render fit for resorption not only *starch*, but also *flesh and other protein-bodies*. Starch (in the form of paste) when introduced into previously cleared and tied loops of gut, was usually converted in the course of three hours into a thin fluid mass, which no longer gave the well-known reaction with iodine. Starch-paste and intestinal juice, when mixed together and exposed to a temperature of from  $35^{\circ}$  to  $40^{\circ}$ , assumed a thin fluid condition in the course of a quarter of an hour, and the mixture was then found to be rich in sugar.

In a similar way pieces of flesh or of coagulated albumen were introduced into tied loops, and in the course of from 6 to 14 hours they were found to be for the most part or entirely digested. It was also shown by experiments, made externally to the organism, that pure alkaline intestinal juice, as well as that secretion when mixed with bile and pancreatic juice, possesses the power of dissolving protein-bodies. Pure intestinal juice dissolved in the course of 6 hours from 36.4 to 40.7% of the flesh digested in it, and very similar ratios were observed when intestinal juice mixed with bile and pancreatic fluid was used. Hence it follows that bile and pancreatic fluid, which impede the digestion of the albuminates by the gastric juice, do not in any way interfere with the digestive powers of the intestinal juice.

We may here refer to a fact which has been previously mentioned namely, that a very large amount of albuminates passes undigested from the stomach, and that the quantity of gastric juice which is secreted is not sufficient to effect the solution of the protein-matter necessary for nutrition; and from this we should obviously conclude that nature has provided some other digestive agent as a solvent for the protein bodies in addition to the gastric juice; and the same remark applies to the saliva and pancreatic juice in relation to the digestion of starch. We have seen that the pancreatic juice disappears, that is to say, is again absorbed before it reaches the middle of the small intestine, and yet we find that starch is readily converted into sugar below this point. These two properties of the intestinal juice are therefore both directly and indirectly proved.

## THE CONTENTS OF THE INTESTINAL CANAL AND THE EXCREMENTS.

The chemical examination of the contents of the intestinal canal has not as yet led to any very certain results; indeed, up to the most recent time, we find that different opinions are held regarding certain points which might easily be decided. We can readily understand the reason of this, when we consider the great variety of matters which must necessarily occur in the intestinal canal. We need hardly observe, that even after tolerably simple food, imperfectly digested and indigestible substances will be simultaneously found in association with already metamorphosed and decomposed matters,\* and that to this already very complicated mixture there are added the constituents of the digestive fluids in every stage of metamorphosis. The difficulty of the investigation lies, however, especially in the circumstance that the digested soluble substances always occur in only extremely minute quantity in those parts of the intestinal canal where they are pretty quickly resorbed. The insoluble substances in the intestinal contents are less accessible to chemical examination, and are unquestionably of less interest in relation to the study of the process of digestion. We shall here limit ourselves to a notice of the actual experiments that have been made on this subject, since the metamorphosis of food as a special process will be subsequently considered when we treat of "Digestion."

In regard to the reaction which the intestinal contents exhibit toward vegetable colors, we may remark, that an acid reaction is always apparent in the duodenum and jejunum; in the ileum it begins to diminish, so that for a great extent before we reach the cæcum, it has often entirely disappeared. As a general rule, the contents of the large intestine are alkaline; it very often, however, happens (as has been previously mentioned) that the inner portions of the contents are still strongly acid, while the outer parts, moistened or permeated with the alkaline intestinal juice, are neutral or alkaline. This acid reaction is usually dependent on the presence of lactic acid, but occasionally on that of butyric, acetic or other acids. The sources of the *lactic acid* are, however, very various, being dependent both on the nature of the food that has been taken, and on the part of the intestine from which the mass has been obtained. In the duodenum, where, notwithstanding the access of bile and pancreatic juice, a strong acid reaction is observed, the free acid depends chiefly on the acid of the gastric juice, whatever kind of food may have been taken; after the use of flesh, sour milk, or acidified food, the acid of the food naturally takes part in the reaction of the contents. In the normal state, it cannot depend on a lactic fermentation, or on any other acid fermentation, since any such fermentation is prevented by the normal gastric juice. On the other hand, it is generally only found in the lower part of the small intestine, and in the large intestine after the use of amylaceous substances; hence, we must conclude that here the reaction is not dependent on the digestive juices, but on the metamorphosed starch. That the free acid which occurs there, is

lactic acid, may be readily proved by analysis (see p. 95). But in the normal condition both starch and sugar are converted in the ileum and the rectum into lactic acid. Moreover, as Frerichs has shown, the lactic acid sometimes becomes transformed into butyric acid in these parts, when all other relations seem perfectly normal. Among the free acids occurring in the small intestine, but exerting less influence on the reaction of its contents, we may mention cholic, glycocholic, and choloidic acids. Frerichs has very thoroughly traced the changes which the biliary constituents undergo in the intestinal canal, and has proved that in the large intestine for the most part we find only dyslysin, but sometimes also a little cholic or choloidic acid.

As a general rule we can, by means of Pettenkofer's test, trace the presence of the resinous constituents of the bile as far as the lower extremity of the ileum (see p. 119.)

Among the less soluble substances which we may extract from the contents of the intestine, we very often meet with *grape-sugar* or *glucose*. This very rarely depends upon sugar having been present in the food; for it is precisely after saccharine food has been taken, that we most rarely find this substance in the small intestine, and then only in its upper part; the sugar introduced into the stomach is unquestionably resorbed from thence, being a readily soluble substance. On the other hand, the sugar found in the small intestine, and sometimes even in the large intestine, owes its origin to the action of the pancreatic juice on starch—an action which, with the co-operation of the intestinal juice, is prolonged to almost the end of the intestinal canal.

In seven cases in which Frerichs fed animals with milk, he could only twice find sugar in the jejunum.

In the aqueous extract of the contents of the small intestine, and occasionally in that obtained from the contents of the large intestine, we find a *protein-body* coagulable by heat, and usually precipitable by acetic acid, always, however, occurring in small quantity. This minute quantity of coagulable matter might well be regarded as a product of the digestion of some protein-body that had been taken as food; for the peptones, which are so readily soluble, are for the most part resorbed from the stomach itself; the digestion of the protein-bodies which pass undissolved from the stomach into the small intestine, cannot be very considerable in the small intestine after the access of the bile. Moreover, the pancreatic juice, to judge by the amount of its secretion, cannot yield any great contribution to the coagulable matter of the aqueous extract of the intestinal contents. But we also invariably find some coagulable albuminous matter after the use of vegetable food poor in protein-bodies, or even of non-nitrogenous food. Hence its sources can only be sought in the exudation of a larger or smaller quantity of albumen from the bloodvessels, in consequence of endosmotic relations.

In four cases in which fasting horses or dogs were fed for two days on balls of starch, and were then killed, I found by no means a very small quantity of coagulable matter in the aqueous extracts of the contents of the jejunum and ileum. In the discharges from the ileum in the above-mentioned case of intestinal fistula, coagulable matter was always found after the use of water-gruel and other slightly nitrogenous

food, and in such quantity that it could not possibly be referred to the protein contained in the bread, groats, &c. We need hardly mention that the precipitate formed on boiling must always be treated with acids and other reagents; for in the watery extract of the intestinal contents, especially in that obtained from the colon, we not unfrequently observe, on heating, a separation which does not depend on albumen, but in part on the relations of weak acid solutions of earthy salts, described in page 303, and in part on the coagulation of mucus, which, if a large quantity of dissolved alkaline salts be present, is very similar to that of albumen. Frerichs has also often found albumen in the colon, and even in the rectum of young dogs and cats after the free use of an animal diet; hence, he inclines to the view, that notwithstanding the impediment which the bile may oppose to the further digestion of the coagulated protein-bodies in the intestinal canal, still, at all events, small quantities of protein-bodies are digested, or at least the modified albumen (peptone) is converted by the bile and pancreatic juice into ordinary albumen. I can by no means assert that this view is erroneous, since it is only by accurate quantitative determinations, which in this case are accompanied with much difficulty, that the point could be decided; but the facts which have been already mentioned, indicate that the coagulable matter which we so frequently meet with in the contents of the intestine, may have its origin in other sources than in the direct conversion of the ingested protein-bodies into soluble and coagulable albumen. I am able in all respects to confirm the results of the experiments of Frerichs, in which he found that soluble albumen was present even in the large intestines of young carnivorous animals, but I attribute it to the presence of undigested flesh; for the contents consisted of lumps of flesh (even when the food had been tolerably finely chopped), and the inner portions of these lumps, reddened litmus, a reaction which might be fairly presumed to depend on the lactic acid originally contained in the flesh. If the alkaline intestinal juices had not neutralized the free acid, the soluble albumen in the flesh would have remained unchanged. It is in the upper part of the small intestine that we find most albumen, because it is there that the contents occur in the most diluted state, and offer the greatest facility for the absorption of albumen from the capillaries.

In the infiltrate of the contents of the small intestines, we only rarely find *dextrin*, and never more than small quantities of *peptones* (Frerichs).

I have never been certain that I have detected dextrin; but there are always to be found small quantities of the substance formerly termed *ptyalin*, soluble in water, but insoluble in alcohol.

If we compare the alcoholic extracts of the different portions of the small and large intestine, we find that *biliary constituents* especially occur in them, in addition to the sugar which has been already mentioned, to the free acids, and to their alkaline salts; and if we treat these alcoholic extracts with ether, we find that this menstruum not only takes up fat, but more or less of certain substances which give the well-known biliary reaction with sugar and sulphuric acid.

That comparatively unchanged bile should be found in the contents of

the duodenum is natural enough, but I have always been struck with the circumstance that biliary substances, and especially the resinous constituents, should be found in the gastric contents of slaughtered animals, and of men that have been suddenly killed. I observed this in a singularly distinct manner in the gastric contents of two horses that had for three days been fed upon starch-balls; the alcoholic extract of the gastric contents was rendered almost as strongly turbid by acetic or hydrochloric acid as that of the duodenal contents; the precipitate, when examined under the microscope, appeared in the form of small vesicular globules grouped together like grapes, which dissolved in boiling water, but resumed their original form as the solution cooled; they readily dissolved in the fixed alkalis and ammonia, as well as in alcohol, but not in ether: the ammoniacal solution, on evaporation, exhibited under the microscope dendritic groupings similar to, but somewhat thicker than those of efflorescent hydrochlorate of ammonia; the potash-solution, on the other hand, yielded crystalline forms resembling the plantain leaf. The solutions of this substance were precipitated by the basic acetate of lead, but not by the neutral acetate or by tannic acid: as it presented the biliary reaction with sugar and sulphuric acid very rapidly and beautifully, it cannot be doubted that unchanged biliary acids—at all events, glycocholic acid—were here present in the stomach as well as in the duodenum.

The further we descend in the intestinal canal, the less of these resinous acids of the bile do we find in the alcoholic extract: but a comparatively larger amount passes into the ethereal extract. Frerichs has also most carefully examined the changes which the bile undergoes in the course of the intestinal canal. We have already remarked that it is chiefly in the small intestine that the presence of the resinous acids of the bile can be easily detected; indeed, near the duodenum we often find bile still undecomposed, which can be recognized in the aqueous extract; the fresh bile discharged into the intestine is very rapidly decomposed by the simultaneous action of the free acid, of the easily metamorphosed protein-bodies, and of the temperature of the animal body; hence we here find only those modifications of the non-nitrogenous choloidic acid which are soluble in alcohol; while further on in the intestinal canal, the greater part of this acid, which is only soluble in alcohol, disappears, and in place of it we find an also gradually diminishing portion of biliary matter soluble in ether, namely, the cholinic and fellic acids of Berzelius, or one of the modifications of Mulder's dyslysin. In the large intestine, and in the solid excrement, I have invariably found a substance soluble only in ether, and in such small quantity that after as correct an estimate as it was possible to institute, it could not be assumed that this was all the bile which had been effused from the liver into the duodenum; but we are rather led to Liebig's view, according to which a large part of the bile is again absorbed in the course of the intestinal canal. As it may be thought that possibly the resinous biliary acids may also be converted into dyslysin which is likewise insoluble in ether, I boiled the contents of the large intestine, and the excrements of men and dogs, after a purely animal diet, with alcohol containing potash; but in the solution which I thus obtained, it was only

rarely that I could recognize biliary resin, that is to say, regenerated choloidic acid, and then only mere traces of it.

I must here again direct attention to the circumstance that in testing the ethereal extract of the intestinal contents for bile, we must go to work with extreme care, lest in employing Pettenkofer's test we confound biliary matter with olein. (See page 481.)

Schmidt propounds the question—to what extent is the bile decomposed in its passage to the middle of the small intestine? In order to decide this question, the quantity of the biliary acids precipitated by acetate of lead was compared with the taurine that is already formed, and which was calculated from the amount of sulphur in the fluid freed from an excess of lead: in 100 parts of the intestinal contents, there were 2.48 parts of fats and biliary acids soluble in ether, 2.021 parts of insoluble biliary matters (cholic, glycocholic, and taurocholic acids), and 0.143 of taurine. Since the latter is equivalent to 0.622 of pure bile-substance, it follows that almost half of the bile effused into the intestinal canal is decomposed before it reaches the middle of the small intestine.

A little *fat* is always found along the whole course of the intestinal canal; and we need hardly observe, that its quantity increases after a fatty diet. After the use of food very rich in fat, we often find such considerable quantities of fat in the solid excrements, that we may obtain a ready confirmation of the results obtained by Boussingault,<sup>1</sup> who found in experiments made on ducks, that in definite times, only certain (not very large) quantities of fat could be resorbed from the intestinal canal. Bidder and Schmidt<sup>2</sup> have recently obtained a precisely similar result in experiments on mammalia. Moreover traces of cholesterin may always be detected in the fat.

[The *fæces* have been submitted to chemical examination during the last few months by Wehsarg,<sup>3</sup> Ihring,<sup>4</sup> and Marcet.<sup>5</sup>—G. E. D.]

The following are the most important points in Wehsarg's Thesis:—The *color* of the normal *fæces* varies with the food; on a mixed diet they are of a yellowish-brown tint, on a flesh-diet they are much darker, and on a milk-diet quite yellow. On exposure to the air the color usually becomes darker, but never red. Very dilute nitric acid, when added in sufficient quantity, always communicates a red color to the *fæces*.

The *odor* almost entirely disappears on drying, or, at all events, becomes less disgusting. It varies with the kind of food. As a general rule the odor is most intense when the stools follow one another rapidly.

The *consistence* seems to depend chiefly on the constitutional relations of the person; but it is considerably influenced by bodily exercise.

The *reaction* is most commonly acid, but not unfrequently alkaline or neutral.

<sup>1</sup> Ann. de Chim. et de Phys. 3 Sér. T. 19, pp. 117–125.

<sup>2</sup> In a private communication.

<sup>3</sup> Mikroskopische und chemische Untersuchungen der Fæces gesunder erwachsener Menschen. Inaug.-Abhandl. Giessen, 1853.

<sup>4</sup> Mikroskopisch-chemische Untersuchungen menschlicher Fæces unter verschiedenen pathologischen Verhältnissen. Inaug.-Abhandl. Giessen, 1853.

<sup>5</sup> Proceedings of the Royal Society, June 15th, 1854. Vol. 7, p. 153.

The number of observations made by Wehsarg was 27; and in 17 of these cases the fæces were those of the 24 hours.

The *quantity* of the daily fæces is very variable; the mean of these 17 observations being 131 grammes (or about 4·6 ounces), the largest and smallest quantities being 306 and 67·2 grammes respectively. This irregularity did not seem in any way connected with an excess of undigested matter. It may be laid down as a general rule, that when the food passes rapidly through the intestine, the daily quantity of the fæces is larger than when it is retained for a longer time in the intestine. In proportion to the rapidity with which the stools follow one another, there is a smaller relative, but larger absolute amount of solid matters. There is no definite relation between the amount of fæces and the bodily weight; the quantity of the fæces seems rather to be connected with the digestive power of the individual.

The fæces, when in a formed or half formed state, contained (taking the mean of 17 observations) 73·3% of water and other matters which were volatile at 120° C., and 26·7% of solid constituents; the latter varied from 17·4 to 31·7%.

The absolute quantity of solid matters discharged in the 24 hours averages 30 grammes, the extremes being 57·2 and 16·3 grammes. No safe inference can be drawn from the consistence of the fæces as to the amount of water and volatile matters that they contain.

The amount of *undigested matters* varies very much in different cases; the mean quantity in 10 observations was 3·4 grammes, or 8·3%, the extremes being 8·2 grammes and 0·81 of a gramme.

A microscopic examination always exhibits remains of the food that has been taken. We commonly meet with vegetable cells and hairs, and spiral vessels in abundant quantity. Muscular fibres colored yellow and corroded by the bile, but still retaining distinct striation, are constantly found. Wehsarg mentions, as of constant occurrence, "a finely comminuted fæcal matter," which appears to be granulo-cellular, but whose structure cannot be distinctly made out; it certainly, however, contains partially destroyed epithelium. Starch is often found. Crystals of ammonia-phosphate of magnesia are always present when the evacuation is neutral or alkaline. Amorphous fat is a constant constituent of the fæces; but Wehsarg never observed crystals of cholesterin: connective tissue was only noticed after a very abundant flesh-diet.

The *ether-extract* of the fæces varied extremely with the nature of the food. After a very fatty diet it rose to 31·2 grammes, or 58·2% of the dried mass; the mean was 11·5%, and the minimum 8·5%. It consists for the most part of a waxy fat.

The *alcohol-extract* was found to amount (as the mean of 3 observations) to 15·6%, and it may rise to double this quantity in diarrhoea. After drying this extract (which when cold forms a dark brownish-red mass) in the air-bath, Wehsarg could only once detect the presence of bile in it with certainty, although he often got doubtful indications; and on the addition of nitric acid to fresh fæces there was only twice an undoubted manifestation of the evidence of bile-pigment. Hence his observations confirm the view, that as a general rule no bile occurs in an unchanged state in the fæces.

The *water-extract* is a brownish-black mass, which always undergoes decomposition on drying. Its average quantity is about 20% of the dry fæces.

The quantity of *salts* contained in the fæces, as compared with that in the urine, is very small. Mere traces of sulphuric acid and chlorine, and often not even a trace, are to be found, unless when large quantities of these substances have been introduced into the system. Chlorine, is, however, more frequently found than sulphuric acid.\*

The salts which are precipitable by ammonia vary in different individuals. The mean of 7 observations was 4·10%, the maximum being 6·90, and the minimum 1·73%. After a dose of sulphate of magnesia, this number may rise to 20·50%. The great mass of these salts is phosphate of magnesia, and associated with it is a small quantity of phosphate of lime with a little iron.

It appears, from Marcet's experiments, that healthy human excrements contain :

1. A new organic substance, possessing an alkaline reaction, which its discoverer names *excretine*. In its pure state it appears in circular groups of crystals, which have the form of acicular four-sided prisms, and polarize light very readily. It is very soluble in ether, cold or hot, but sparingly soluble in cold alcohol ; it is insoluble in water, and is not decomposed by dilute mineral acids. It fuses between 95° and 96° C., and at a higher temperature burns away without inorganic residue. It does not dissolve when boiled with a solution of potash. It contains nitrogen and sulphur, though in small proportions. The products of its decomposition have not yet been investigated. Marcet considers that it exists for the most part in a free state in the excrements, and constitutes one of their immediate principles. As to its source, he observes that it appeared in excess when a considerable quantity of beef had been taken, and in less than the usual quantity in a case of diarrhœa attended with loss of appetite ; but none could be directly obtained from beef on subjecting it to the same process of extraction as fæces ; neither could it be found in ox-bile, the urine, or the substance of the spleen.\*

2. A fatty acid having the properties of margaric acid, but not constantly present. He is uncertain whether the margaric acid in the fæces is free, or combined with excretine, but he is disposed to conclude that the neutral fats are decomposed in the intestinal canal, and their acid set free. Not having been able to discover stearic acid in human evacuations, he supposes that what is contained in the fat taken in the food must be converted into margaric acid in its passage through the alimentary canal.

3. A coloring matter similar to that of blood and urine.

4. A light granular substance, which he is inclined to regard as a combination of phosphate of potash, and a pure organic matter.

5. An acid olive-colored substance, of a fatty nature, which he names *excretolic acid*. It fuses between 25° and 26° C., and at a higher temperature burns without residue. It is insoluble in water, and in a boiling solution of potash, is very soluble in ether, and in hot alcohol, and slightly so in cold water. He believes that it is combined in the excrements in the form of salt with excretine or a basic substance closely allied to it.



6. No evidence of butyric or of lactic acid was obtained.

The fæces of various animals yielded the following results :

1. The excrements of carnivorous mammals, viz., the tiger, leopard, and dog (fed on meat), contain a substance allied in its nature to excretine, but not identical with it. They contain no excretine, but yield butyric acid, which is not present in human excrements.

2. The excrements of the crocodile contain cholesterin, and no uric acid, while those of the boa yield uric acid, and no cholesterin. [It is probable that the semi-solid urine and the excrements were not duly separated in this experiment.—G. E. D.]

3. The fæces of herbivorous animals, viz., the horse, sheep, dog (fed on bread), wild boar, elephant, deer, and monkey, contain no excretine, no butyric acid, and no cholesterin.

Ihring has examined the evacuations after the use of chloride of sodium, Nauheimer water, and of preparations of iron, and in cases of intestinal tuberculosis, bilious diarrhoea, &c., and has likewise submitted to investigation the contents of different parts of the intestinal contents in a patient who died from a chronic affection of the stomach. [We must refer to his thesis for further particulars.—G. E. D.]

The *bile-pigment* also gradually undergoes the same changes in the intestinal canal as are observed to occur in the putrefaction or decomposition of the bile. It is only in the alcoholic, and occasionally in the aqueous extract of the contents of the small intestine, that we can induce the well-known changes of color by the mixture of sulphuric and nitric acids; in the large intestine, the bile-pigments in all probability occur under the same modification, which, according to Berzelius and Scherer, is to be regarded as the final product of the metamorphosis of cholepyrrhin.

*Taurin* has often, although not invariably, been detected by Frerichs<sup>1</sup> in the whole course of the intestinal canal, and even in the solid excrements.

The *constituents* of the intestinal canal *insoluble* in water, alcohol, and ether, fall, for the most part, within the domain of microscopic inquiry. They essentially consist of undigested or indigestible fragments of food. Among the undigested substances we commonly find not only fat-globules, but starch-granules, fibres of muscle, and fibrils of cellular (areolar) tissue in the excrements after the use of the corresponding articles of food. The *starch-granules* seem to be diminished in their diameter, and this diminution is the more marked the lower they are found in the intestinal canal; they usually appear fissured and lobulated, and as if some of their coats were partly or entirely dissolved; in this case their true nature can often not be detected under the microscope, unless with the aid of a solution of iodine. *Muscular fibres* are found in every phase of change; we recognize some primitive fibres unchanged in their histological formation, and parallelipeds of the same structure, in which the striæ may be pretty clearly made out, presenting a finely punctated appearance; the longitudinal striæ are usually the most distinct; the sarcolemma has, for the most part, disappeared; finally, there often remains merely a tolerably hyaline mass, which can

<sup>1</sup> Op. cit. p. 841.

only be recognized as the remains of muscular fibre by the parallel grouping of a few prominent points. A complete solution of muscular fibre is not effected by the gastric and other digestive juices, as has also been found by Frerichs.

*Fragments of bone*, after being swallowed, may be always detected in the intestine and in the excrements, although a great part of them is obviously dissolved in the *primæ viæ*.

As the *histological constituents of the vegetable tissues* have the least tendency to be decomposed by the digestive juices, they are always found comparatively little changed after the use of vegetable food; cellulose is proof against all organic solvents, and hence we meet with all varieties of vegetable cells. The chlorophylle-cells remain unchanged; the parenchyma-cells are only sometimes isolated; spiral vessels may be beautifully seen in the excrements both of the higher and the lower animals. Yeast-cells are often met with after the use of pastry.

In addition to the fluid and solid contents of the intestinal canal, we must also refer to the *gases* occurring there. Unfortunately, however, the very few observations which we possess regarding these elastic fluids are not altogether trustworthy, since the investigations made regarding the gas contained in the intestines in cases of disease, have usually not been instituted till twenty-four hours after death. Magendie and Chevreul<sup>1</sup> are the only experimentalists who have examined the gaseous contents of the stomach and the small and large intestines of men immediately after their execution; and even these investigations cannot be regarded as altogether conclusive, since a person's knowledge, that he is going to be executed in a few hours, must probably somewhat disturb his digestive functions.

In the stomach of a man, after execution, Magendie and Chevreul found a gaseous mixture, consisting of atmospheric air, in which a portion of the oxygen had been replaced by carbonic acid; and, besides this, they found a little hydrogen. (According to volume, this air was composed of 14% of carbonic acid, 11% of oxygen, 71·45% of nitrogen, and 3·55% of hydrogen.) Moreover, it can hardly be doubted that this air was for the most part conveyed into the stomach from without. We have already mentioned that, in the insalivation of the food, a very appreciable quantity of air is mixed with it, and this is probably the most common mode by which atmospheric air finds its way into the stomach, although, in certain respiratory movements some air may be driven or pressed through the œsophagus, as, for instance, in the efforts which precede vomiting, as has been shown by Budge: some persons, however, possess the power of swallowing air at will, and of exciting vomiting by swallowing large quantities.

The diminution of the oxygen, and the considerable augmentation of the carbonic acid, may be referred with more probability to the interchange of these gases with those of the blood, than to processes of fermentation; this interchange is, at all events, a physical necessity, while processes of fermentation are always indicative of something abnormal in the stomach. In the case examined by Magendie and Chevreul, there

<sup>1</sup> Berzelius, *Lehrb. d. Ch.* Bd. 9, S. 338-340.

certainly seems to have been a fermentation, as evidenced by the presence of hydrogen, although in small quantity, in the air.

In the dead bodies of healthy men and animals, the quantity of air found in the stomach is always extremely small; but there are various conditions in which there is an abnormal accumulation of air in the stomach, and some have even regarded this symptom as a special disease, and have termed it *pneumatosis ventriculi*. Even in healthy persons, large quantities of gas may accumulate in the stomach after the use of such kinds of food and drink as very readily undergo fermentation, as, for instance, biscuits rich in yeast, new bread, onions, garlic, radishes, raw fruit, or imperfectly fermented wine and beer, especially when taken in very large quantities. In such cases, a great excess of carbonic acid is always found in the stomach, since all these substances undergo the vinous and acetous fermentation, which is almost always preceded by the development of carbonic acid. If, however, hydrogen gas be found to occur in this air, its presence may be easily explained, since, as we have already seen, the amylacea have a strong tendency to undergo the butyric fermentation in the stomach, and this fermentation is always accompanied, as has been shown by Pelouze, Liebig, and others, by the development of hydrogen.

Accumulations of air in the stomach are especially observed in hysterical and hypochondriacal patients, who have an unnatural tendency to gulp air, in persons in whom the food is retained for too long a period in the stomach, and finally, in cases in which the secretion of the gastric juice is altogether impeded. In hysterical and hypochondriacal patients who have swallowed air, the gases evolved by eructation are, for the most part, devoid of odor, and hence it may be presumed that this air has undergone very little change, except an augmentation of carbonic acid.

In constrictions of the pylorus, as well as in chronic catarrhs, the stomach becomes filled with air, not only after the moderate use of the above-mentioned articles of diet, but also after the ingestion of other varieties of food which do not usually cause any annoyance to healthy persons, or at the most only occasion accumulations of gas in the large intestine, as, for instance, milk, peas, cabbage, eggs, meat, and other animal food. In such cases the air contains only little oxygen, much carbonic acid, probably also hydrogen and carburetted hydrogen, and invariably sulphuretted hydrogen, which may be recognized by the smell of the eructations, as well as by its reaction on paper moistened with a solution of acetate of lead.

In patients suffering from typhus fever, who for a considerable time have taken neither food nor medicine, the stomach is not unfrequently found to be distended with gas: here the meteorism only comes on slowly, and its occurrence is very much favored by the paralytic condition of the muscular coat of the stomach.

Chevillot<sup>1</sup> found from 25.2 to 27.8% (by volume) of carbonic acid, from 8.2 to 13.0% of oxygen, and from 66.8 to 59.2% of nitrogen, with mere

<sup>1</sup> Journ. de Chim. Méd. 1 Sér. T. 5, p. 596-650, and Arch. gén. de Méd. 2 Sér. T. 5, p. 285-292.

traces of hydrogen, in gas taken from the stomach twenty-four hours after death.

In the *small intestine* we usually find far less gas than in the large intestine: in the small intestines of three persons who had been executed, Magendie and Chevreul found no oxygen, but an extraordinary abundance of hydrogen and carbonic acid (in the first case 24.39%  $\text{CO}_2$ , 20.08% N, and 55.53% H; in the second case, 40.00%  $\text{CO}_2$ , 8.85% N, and 51.15% H; and in the third case, 25.0%  $\text{CO}_2$ , 66.6% N, and 8.4% H); Chevallot,<sup>1</sup> on the other hand, always found 2 or 3% of oxygen in the air discharged from the small intestines of the bodies of aged persons. We can easily understand how in cases of disease, and even in healthy persons, after the use of flatulent food or drink, these accumulations of gas occur more frequently than in the stomach; for on the one hand, the flatus is not so readily discharged from hence by eructation as from the stomach, and on the other, the fermentation and decomposition of the above-named substances proceed here with a rapidity proportional to the length of time they have already remained in the stomach and small intestine. Constrictions of individual portions of the small intestine, and other diseases of the intestinal tube, contribute also essentially to the augmentation of these accumulations of gas.

On comparing the composition of the air from the small intestine with that of the gas obtained from the stomach, we observe in the one case a perfectly opposite relation to that which holds good in the other; we have here to deal with mere residual traces of atmospheric air, the greater portion of the gas having its source in the decomposition of nitrogenous and non-nitrogenous substances. We must, however, always bear in mind that these gases are only separated from those of the blood by permeable moist membranes, and that, for this reason, the analysis of the air never correctly expresses the gaseous products arising from the decomposition of the food. Hence it is more than probable that the symptoms of meteorism, which in children and hysterical women occasionally supervene to a dangerous extent, are not merely dependent on the mechanical contraction of the thoracic cavity (by the upward pressure of the diaphragm), but also on the transmission of certain gases into the blood. In these cases we should not so much suspect the resorption of carbonic acid as of hydrogen and its compounds. The amy-lacea, in undergoing butyric fermentation, which is only impeded in the intestine by the free acid of the gastric juice, yield hydrogen, which in its nascent state unites with the sulphur of the decomposed protein-bodies, and thus produces the sulphuretted hydrogen, which exerts so injurious an effect on the blood. The presence of sulphuretted hydrogen in the gaseous contents of the small intestine may, moreover, be readily perceived from the eructations which are developed in from four to eight hours after a meal. It is further worthy of notice, that these eructations of sulphuretted hydrogen are very common after the use of ferruginous preparations; it is possible that the presence of iron facilitates the conversion of the alkaline sulphates into metallic sulphides,

<sup>1</sup> [On referring to the Journ. de Chim. Méd., we find that oxygen was only found in the small intestines *once* in fifty-four cases; in that case the proportion was from 2 to 3%.—G. E. D.]

and occasions the formation of sulphide of iron, whose decomposition by acids gives rise to the production of sulphuretted hydrogen. The formation of sulphuretted hydrogen after the use of the preparations of sulphur, is so well known an occurrence as hardly to require notice, and demands no explanation.

Gaseous accumulations are much more frequent in the large intestine, where they are often very considerable, than in the stomach and small intestine. According to Magendie and Chevreul's investigations, the oxygen has here altogether disappeared; they found from 43.5 to 70% of carbonic acid, from 18.40 to 51.03% of nitrogen, and from 5.47 to 11.6% of carburetted hydrogen: Chevillot<sup>1</sup> found in the gas contained in the large intestines of aged persons, from 23.11 to 93.00% of carbonic acid, from 2 to 3% of oxygen, from 95.2 to 90.0% of nitrogen, and 28.0% of carburetted hydrogen. In two analyses of the flatus, Marchand found 36.5 and 44.5% of carbonic acid, 29.0 and 14.0% of nitrogen, 13.5 and 15.8% of hydrogen, 22.0 and 15.5% of carburetted hydrogen, and in the latter of the cases 1.0% of sulphuretted hydrogen. It is worthy of remark that the sulphuretted hydrogen always occurs in the gases of the large intestine in far less quantity than we should have expected from the odor. It is hardly necessary to indicate the reasons why the development of gas is always more considerable in the large than in the small intestine; for although the decomposition of the remains of the food may have begun in the ileum, it proceeds with greater rapidity in the colon, since there the fecal mass no longer meets with any free acid to impede its further decomposition. Should, however, the contents of the large intestine be acid, this, as we have already shown, must depend on a butyric fermentation, which indeed is accompanied by a copious development of gas. We need not trouble the rational physician with a detailed notice of all those morbid conditions which lead to large accumulations of air in the cæcum and the colon; it is sufficient for us simply to mention that these accumulations of gas, which we are accustomed to term meteorism or flatulence, may either be a consequence of suppressed or perverse secretion of the intestinal juices, or of diminished contractility of the muscular coat of the intestine, of strictures and other anatomical changes of the colon, of pressure exerted by morbid tumors on the lower parts of the intestine, &c. Substances stagnating in the different parts of the colon, undergo complete putrefaction, and their products, gaseous as well as solid, are precisely the same as we observe out of the animal body. Thus, in the examination of such masses, Frerichs found substances precisely similar to those which Bopp has obtained from putrefying protein-bodies.

The early physicians believed in a secretion of gas from the walls of the intestine; to those who are at all acquainted with the law of the

<sup>1</sup> [On referring to the Journ. de Chim. Méd., we find that the largest quantity of carbonic acid discovered in the digestive canal generally was from 92 to 93%, and that the mean quantity in the large intestines was 23.11%. The quantity of oxygen is not stated: Chevillot only observes that he found it in the large intestine five times in fifty-four cases. The mean quantity of nitrogen in the large intestines of twenty-seven aged persons was 73%; the maximum is not given in the memoir. In ninety-six cases, ten only afforded carburetted hydrogen; one in the small intestine, and nine in the large intestine. The greatest quantity found was 18.8%.—G. E. D.]

metamorphosis of the animal tissues and with the chemical processes of putrefaction, such an assumption is altogether unnecessary for the explanation of considerable accumulations of gas; and further, from what is known on the subject, it is very improbable that gases, such as hydrogen, carburetted hydrogen, and sulphuretted hydrogen (which latter we do not find in the blood), should pass from the general juices of the body into the intestinal canal. Magendie and Girardin<sup>1</sup> have, however, made an observation which has also been confirmed by Frerichs,<sup>2</sup> which, at all events, proves the possibility of a secretion of gas from the blood into the intestine; for if a loop of intestine in dogs, after being perfectly emptied of its contents, were tied at both ends, it was always found after some time to be filled with air. It is to be regretted that this air has not been analyzed; it is scarcely likely that hydrogen and its gaseous compounds would be found in it.

Frerichs likewise notices an accumulation of gas, which, strictly speaking, is a sacculated emphysema in the serous coat of the gut; in the intestines of swine he has frequently observed bullæ of this sort, as large as a pea or a hazel-nut, filled with air.

Although from what has been already stated it may be readily inferred what are the substances which occur, and which must of necessity occur, in the matters discharged by vomiting, it yet may not be altogether superfluous to notice systematically the different characters of the vomitus in different conditions of disease. Unfortunately, many of the analyses which have been made are of little use: as in the diagnosis of a gastric disease, so also for a scientific investigation of vomited matter, it is especially important to know what period had elapsed since food was taken, or whether the stomach was empty. Without this knowledge no inference of any scientific value can be deduced. It is, however, much to be lamented that even at the present day pathological chemistry (as it is called) is as little based on physical diagnosis as on pathological anatomy; thus we find numerous analyses of the vomitus in dyspepsia,—a word unsatisfactory to every rational physician, and tending only to impede scientific inquiry. Every one must know that dyspepsia and pyrosis may accompany not only chronic gastric catarrh, but also the round (perforating) ulcer, cancer, and other primary and secondary affections of the stomach; if then no pathologico-anatomical diagnosis be made, the analysis of the matters vomited by dyspeptic patients can lead to no result; when it is impossible to make a certain diagnosis in dyspepsia or pyrosis, nothing is gained by the attempt to analyze the vomited matters. Notwithstanding the numerous, more or less accurate analyses of vomited matters, we still know very little regarding the various morphological and chemical constituents of the masses which are discharged in the various diseases of the stomach and other abdominal organs. All that is positively known may be included in a few sentences.

By far the most frequent cases are those in which the principal part of the vomited matter consists of imperfectly digested or entirely undigested food, and the chief reason of this is that the food is usually the

<sup>1</sup> Recherches physiol. sur les Gaz. intestin. Paris, 1824, p. 24.

<sup>2</sup> Op. cit. p. 866.

proximate exciting cause of the antiperistaltic motion. Hence it follows that the food is more or less changed according to the time in which it has been retained in the stomach: thus in the round ulcer of the duodenum where vomiting occurs four or six hours after food has been taken, we constantly find that not only the albuminous substances, but also the amylacea, are far more changed than in perforating ulcer of the stomach; in scirrhus of the pylorus, on the other hand, they are usually less changed than in other cancerous affections of the stomach, &c. These changes which we perceive in the food must either be normal or abnormal, that is to say, in the first case we find half-digested muscular fibre, peptones, sugar, &c., changed in the manner which has been already described. These are the rarer cases, and for the most part occur when the seat of the disease which has occasioned the vomiting lies externally to the stomach, although sometimes also in cancer of the stomach. It far more frequently happens that the food, when it has remained for a prolonged period in the stomach, has undergone abnormal changes; if saccharine or amylaceous food has been taken, lactic, acetic, or butyric fermentation is induced, in which case the vomited matters have an extremely strong acid reaction and taste, and even seem to take the edge off the teeth; the nitrogenous articles of food appear in this case, when examined under the microscope, to be but slightly changed, and at most to be only loosened in texture and rendered more transparent; matters of this nature are principally vomited in chronic gastric catarrh, but not unfrequently also in round (perforating) ulcer and in cancer of the stomach. It seems probable that in chronic catarrh of the stomach, all those kinds of fermentation may be set up in the starch, according to the nature of the secreted mucus, which we are accustomed to observe out of the animal body in the laboratory of the chemist, just as in catarrh of the urinary bladder there is sometimes a predisposition to acid and sometimes to alkaline urinary fermentation. Certain experiments made by Frerichs show that in diabetic patients there is a special tendency to the formation of sugar in the stomach. Another of his observations is even more important; he convinced himself that the colorless, viscid, ropy masses, which are sometimes ejected in abundant quantity in gastric catarrh, possess almost entirely the same properties as the gun-like substances produced by what is called mucous fermentation. It appears to depend, at all events in part, on the nature of the mucus secreted in gastric catarrh, whether the fermentation established in the amylacea be of the mucous, lactic, acetic, or butyric variety—a view which seems to correspond with our present knowledge of the exciters of these different kinds of fermentation, and with the different anatomical changes of the gastric mucous membrane and of the mucus secreted by it.

Masses in a thoroughly digested state, and at the same time in an almost putrid condition, are only vomited in cases of some anatomico-mechanical change in the intestinal canal, as strangulated hernia, volvulus, &c. Since, as we have already mentioned, yeast-fungi are sometimes found in the contents of the stomach and intestines—partly entering from without, and partly propagated within the body—it need excite no wonder that they are also found in vomited matters. The same may be said regarding the *sarcina*, whose nature and mode of occurrence, since

its discovery by Goodsir,<sup>1</sup> have given rise to so many investigations and discussions. This organized being is in all probability identical with the alga, *Merismopedia punctata*, that had been described by Meyen,<sup>2</sup> and with the *Gonium tranquillum et glaucum*, referred by Ehrenberg<sup>3</sup> to the *Bacillariæ*; it forms smooth plates, consisting of a larger or smaller number of quadrupartite cells, which range from 1-300th to 1-500th of a line in diameter, are square, and resemble tied-up packets; these may be found singly in the vomited matters, but much more frequently hanging together in regular forms in fours, eights, and sixteens, so as to form larger surfaces. These algæ are not characteristic of any special disease of the stomach, either organic or functional, although they are most commonly found when the food has been retained for a considerable time in the stomach before the vomiting has occurred, as, for instance, in cancer of the stomach. Frerichs<sup>4</sup> has frequently found the sarcina in the stomach after death, in cases in which, during life, no signs of deranged digestion had been observed; indeed, he even noticed it in a dog with a gastric fistula, and found that the digestion went on as regularly and energetically as before the appearance of these algæ. It thus appears to have no connection with any pathological phenomena in the animal organism.

Hence the sarcina is of no diagnostic value, since neither its production nor its growth is dependent upon, or gives rise to any special morbid processes.

Frerichs has studied its development in a dog with a gastric fistula; he observed, first of all, round non-nucleated cells, generally isolated, but sometimes grouped two and two, and ranging from 1-400th to 1-300th of a line. The cell, which at first is transparent, gradually undergoes a superficial constriction through its centre, and this is crossed by a similar constriction at right angles; the lines deepen from the centre towards the periphery, till, finally, the cells appear to be divided into four equal parts, the separate squares ranging from 1-700th to 1-500th of a line: as each of these squares again subdivides in the same manner into four fresh squares, the original individual expands into large plates, which are intersected by rectangular lines, and are easily broken down into separate quadrupartite cells.

Hasse has also found the sarcina in evacuations from the bowels; and Heller<sup>5</sup> appears to have found it in a urinary sediment, although he does not seem certain of its identity.

Hasse and Kölliker,<sup>6</sup> Virchow,<sup>7</sup> and more especially Schlossberger,<sup>8</sup> have instituted accurate chemical inquiries regarding the constitution of this body. Virchow found that the molecules of the sarcina were not changed by acetic acid, but that potash first rendered them more transparent, and subsequently caused their disintegration into amorphous granules. Hasse and Kölliker found that acids and alkalies only rendered the sarcina paler; that it dissolved when boiled in sulphuric acid; that when boiled with hydrochloric acid the larger parts separated into

<sup>1</sup> Edinburgh Med. and Surg. Journal. Vol. 57, p. 430.

<sup>2</sup> Neues System der Pflanzen. Bd. 6, S. 410.

<sup>3</sup> Infusorien, S. 58, Taf. 8, Fig. 3.

<sup>4</sup> Häser's Arch. Bd. 10, S. 175-208.

<sup>5</sup> Arch. f. phys. u. path. Chem. Bd. 4, S. 308, Taf. 1, fig. 5.

<sup>6</sup> Mittheil. der Zürcher naturf. Gesellsch. 1847. S. 95.

<sup>7</sup> Arch. f. path. Anat. Bd. 1, S. 364.

<sup>8</sup> Arch. f. phys. Heilk. Bd. 6, S. 747-768.



smaller; that in a hot solution of potash the contents partially dissolved, leaving a perfect skeleton; and finally, that the sarcina, after being treated with sulphuric acid, was only colored yellow by iodine, but that at a glowing heat, it was perfectly destroyed. The conclusions of Schlossberger were, that the sarcina was unaffected by water, alcohol, ether, and the fats as well as the volatile oils, and that neither organic nor dilute mineral acids apparently acted on it. When treated with iodine and sulphuric acid, in order to test for cellulose (according to Mulder's method), it exhibited no blue or greenish color; concentrated sulphuric acid decolorized the sarcina, and rendered it very transparent; the interspaces between the greatest squares became swollen, and on the addition of water, the larger broke into smaller parts. When the action was prolonged, it entirely dissolved; many were rendered yellow by nitric acid, only, however, when they had been previously treated with a solution of potash; hence they appeared, at all events, in part to contain a protein-like constituent. But, on the other hand, Schlossberger could not obtain a blue color with hydrochloric acid; indeed, he expresses great doubts whether there is any difference between the capsule and the contents, although Hasse and Kölliker believe that they had proved, both by hydrochloric acid and by potash, that a difference existed. Caustic potash causes the sarcina, or at all events its larger interstices, to swell. The sarcina is unaffected by alcoholic and acid fermentation.

Far more amenable to chemical investigation, and of more physiological interest, are the (generally) fluid materials which are sometimes vomited in the *fasting* state; as, for instance, in chronic catarrh of the stomach, in the round (perforating) ulcer, and in cancer of the stomach. Although the investigation of such secretions is indispensable to a right comprehension of the nature of the substances which, mixed with food, are usually vomited, we have as yet only few analyses of these gastric and intestinal secretions discharged by the mouth, and still fewer in which the diagnosis of the disease has been established. Thus, for instance, waterbrash (pyrosis) has excited the attention of physicians, and the vomited matter has been analyzed, and, on one occasion, the fluid has been found alkaline, and on another strongly acid, without any regard to the pathologico-chemical process. Frerichs<sup>1</sup> has here also opened the path for further inquiry; he has ascertained that, in many forms of gastric disease, as, for instance, in the chronic gastric catarrh of drunkards, and sometimes in cancer and round (perforating) ulcer of the stomach, the salivary glands are consensually irritated, and secrete an abundance of saliva, which accumulates in the stomach, and finally induces vomiting. In such cases the vomited fluids present all the characters of saliva; they are in most cases alkaline, often however neutral, rarely acid, contained a large quantity of the sulphocyanides, and, under the requisite conditions, converted starch very rapidly into sugar.

These fluids were found by Frerichs to be slightly turbid in consequence of the presence of epithelium and fat-globules; their density varied from 1.004 to 1.007, and they contained from 0.472 to 0.6888 of solid constituents; the application of heat did not much increase their turbidity; the addition of alcohol caused a separation of white flocculi,

<sup>1</sup> Op. cit.

which possessed the metamorphic power on starch in a high degree; the watery solution of their alcoholic extracts assumed a dark blood-red tint with the per-salts of iron. Similar kinds of alkaline vomited fluids have been examined by Wright, Nasse,<sup>1</sup> and Bird.<sup>2</sup>

We very often observe a fluid, watery, vomited matter with a strong acid reaction; it occurs in the round (perforating) gastric ulcers and probably also in nervous spasm of the stomach (if such a thing actually exist). Unfortunately these fluids have been examined with so little care that even if lactic, butyric, or acetic acid has been actually recognized in them with chemical certainty, it has not been decided whether the excess of acid is produced in the same way as in softening of the stomach in children (Elsässer) by the rapid fermentation of portions of amylaceous or sugar-forming food retained in the stomach, or whether it has accumulated in the stomach in consequence of an abnormal secretion from the gastric glands.

The fluids of this class that have been most frequently analyzed are the *rice-water* matters vomited in *cholera*; both in their physical and in their chemical properties they are almost perfectly identical with the matters often vomited in *uræmia*; they are usually of a faint, sickly odor, and their reaction may be either acid, neutral or alkaline; on standing, they deposit grayish-white flakes, consisting of epithelial structures or intestinal mucus, while the fluid above appears clear and yellowish. With the exception of very beautiful groups of cylindrical epithelium, we find in these fluids only few organic matters; but, on the other hand, they contain a relatively large amount of inorganic salts, and especially of chloride of sodium, with a small quantity of alkaline sulphates. It entirely depends upon the stage of the disease whether the fluid is acid or alkaline; for a short time after the beginning of the disease the vomited matter is acid, and I found in it (as Hermann<sup>3</sup> had done) butyric and acetic acids (and metacetic acid was also very probably present). When the fluid contained no remains of food, but resembled rice-water, and was acid or neutral, I constantly found *urea*, and can thus confirm the observations of Schmidt.<sup>4</sup> If, on the other hand, the disease was further advanced, and the cerebral symptoms accompanying *uræmia* had set in, and if vomiting now came on, salts of ammonia, and especially the carbonate, were found, and hence the fluid had an alkaline reaction. *Albumen* occurs only in very small quantities when the fluid is acid, but in larger quantities when there is an alkaline reaction.

The specific gravity of these fluids varies from 1.025 to 1.007; they contain from 0.4 to 0.6% of solid constituents, of which more than half are often inorganic (Wittstock,<sup>5</sup> Mulder,<sup>6</sup> Andral,<sup>7</sup> A. Taylor,<sup>8</sup> Becquerel,<sup>9</sup> Guterbock,<sup>10</sup> Schmidt).

The albumen, regarding whose presence or absence in the cholera-

<sup>1</sup> Med. Correspondenzbl. rh. u. westph. Aerzte, 1844, No. 14.

<sup>2</sup> Lond. Med. Gaz. Vol. 29, p. 378, and Vol. 30, p. 931.

<sup>3</sup> Pogg. Ann. Bd. 22, S. 169.

<sup>4</sup> Charakteristik der Cholera, u. s. w. S. 72.

<sup>5</sup> Pogg. Ann. Bd. 24, S. 625.

<sup>6</sup> Natuur en Scheikundig Archief. D. 1, st. 1, 1833.

<sup>7</sup> Gaz. Méd. 1847, p. 654.

<sup>8</sup> Chem. Gaz. 1849, p. 95.

<sup>9</sup> Arch. gén. de Méd. 4 Sér. T. 21, p. 192.

<sup>10</sup> Journ. f. pr. Ch. Bd. 48, S. 780, u. 850.

dejections there has been so much discussion, can generally only be recognized by the aid of hydrochlorate of ammonia, or, if the reaction of the fluid be alkaline, by its neutralization.

*Biliary matters* are contained in the vomited matters under very different conditions. We most commonly find biliary matters vomited simultaneously with the remains of food; and, by a careful chemical examination, the biliary acids may be detected by Pettenkofer's test in most substances discharged by vomiting; it is also easy to understand how the contents of the small intestine, including the constituents of the bile, are ejected by antiperistaltic motion. We meet with larger quantities of bile mixed with slight remnants of food or only with gastric juice and saliva, in the matters vomited in inflammatory conditions of the abdominal organs, especially of the peritoneum, as well as in cerebral affections of an inflammatory nature; the vomited matter is then of a grass-green, or verdigris color (*vomitus æruginosus*). The green color of these fluids is dependent on the green modification of the bile-pigment, which is induced by the action of the free acid of the gastric juice on the brown pigment: the fluid has generally a strong acid reaction, and on the addition of sulphuric with an admixture of nitric acid, or of the latter acid alone, exhibits the most beautiful changes of color peculiar to the bile-pigment. It usually contains no substance coagulable by heat, but saliva is present, as, at least, may be inferred from the circumstance that sulphocyanides may be detected in the alcoholic extract. As in all vomited matters, we here find pavement and cylindrical epithelium and fat-globules, in addition to saliva; the fat-globules in this case, when examined under the microscope, usually exhibit a green color, from the presence of cholepyrrhin.

*Bloody vomiting* may, as is well known, be associated with very various conditions. The blood is often still fluid and of a tolerably bright color when it is ejected very soon after its escape from the vessels; but most commonly it is of a dark brown red color, coagulated, and mixed with fragments of food. In capillary gastric hemorrhage, which may take its origin in various diseased conditions, as, for instance, in round (perforating) ulcer of the stomach, in gastric cancer, in hemorrhagic erosions of the mucous membrane of the stomach, and in disturbances in the circulation in the spleen and liver, the blood is retained for a longer time in the stomach, and we then have the brown or black vomitus, having the color of chocolate or resembling coffee-grounds, to which the earlier pathologists attached so much importance. The remains of blood-corpuscles are always to be found on examining this kind of vomitus with the microscope. Any one not trusting to his powers as a microscopist, may easily obtain a red fluid by heating the dried mass with alcohol containing sulphuric acid, in which the presence of hæmatin is indicated not merely by the general character of its solid residue, but also by the abundance of iron in the latter. Fat-globules, epithelial structures, &c., are also found in these masses.

*Sugar* has very often been found in vomited matters: MacGregor,<sup>1</sup> Polli,<sup>2</sup> and, more recently, Scharlau,<sup>3</sup> have found it in the contents of the stomachs of diabetic patients: two observations made by Frerichs,<sup>4</sup> appear

<sup>1</sup> Lond. Med. Gaz. May, 1837.

<sup>2</sup> Zuckerharnruhr, Berlin, 1846.

<sup>3</sup> Omodei annali univers., 1839.

<sup>4</sup> Op. cit. p. 804.

to confirm these observations; for in the matters vomited by diabetic patients after the administration of an emetic, he found a large quantity of sugar but no dextrin. It was also worthy of remark that notwithstanding the neutralization of the acid vomited matters, no lactic fermentation could be induced.

Frerichs believes that this experiment throws some light on the pathogenesis of diabetes. Although this indication, if it turn out to be constantly exhibited, should undoubtedly not be overlooked, yet we still think that the proximate and essential cause of diabetes is hardly to be sought in the *primæ viæ*, for in the normal condition, starch is converted into sugar in the stomach, and sugar is found in the blood; moreover, sugar is formed in the liver; that is to say, it is not only found therein, as Bernard asserts, but, as I have observed, far more sugar proceeds from the liver through the hepatic veins, than is conveyed to it through the portal vein and the hepatic artery; hence, for the present, it seems more correct to assume with C. Schmidt, that in diabetes the conversion or regressive formation of the sugar is impeded. Moreover, it need not excite our wonder that a large quantity of sugar is found in the contents of the stomachs of diabetic patients, since there is no improbability in the supposition that sugar is also separated by the gastric glands as well as by the salivary glands from the diabetic blood.

Nasse<sup>1</sup> has observed a remarkable case in which large quantities of fat were vomited. No evidence could be adduced to show that the fat had been introduced from without into the stomach.\*

Although the general character of the *solid excrements* in the normal state must be sufficiently obvious, from the above sketch of the changes which the individual substances undergo in the intestinal canal till they reach the rectum, we must return to the subject for the purpose of considering the pathological relations of the intestinal excretions. Important as is the investigation of this subject for physiologists, and especially for physicians, our investigations regarding it are as yet few and of doubtful accuracy. The analysis of the solid excrements is, however, attended with so many difficulties, and is so disgusting a task, that we find it exciting the complaints even of a Berzelius. Putting out of the question the repugnance which must be overcome before we can handle and apply heat to such matters, the extreme varieties which the excrements present according to the nature of the food that has been taken, and the great facility with which decomposition extends in such masses, we are hindered from making a tolerably correct analysis, by the circumstance that all solutions pass in a turbid state through the filter, and that the decomposed biliary constituents distribute themselves through all menstrua, so that we cannot readily extract a substance to which some decomposed bile-pigment or putrid biliary matter does not adhere.

An adult male, in a state of health, living on a mixed diet, usually discharges in the course of twenty-four hours from 120 to 180 grammes of semi-solid brown masses, whose unpleasant odor seems from Valentin's experiments to be far more dependent on decomposed constituents of the bile than on the remains of the food. These masses contain about

<sup>1</sup> Med. Correspondenzbl. rh. u. westph. Aerzte, 1844, Nr. 14.

25g of solid constituents, so that from 30 to 45 grammes of solid dry matter are daily carried off in the intestinal evacuations of a healthy man living on a mixed diet.

As, in our remarks on the contents of the large intestine, we have at the same time considered the constituents of the fæces, we now proceed to point out the differences which the excrements present under special physiological and pathological conditions.

It is almost unnecessary to introduce the remark that indigestible fragments of food, as vegetable cellular tissue, tendons, skin, &c., occur in the fæces in varying quantities according to the nature of the food, and that the amount of undecomposed bile which is found, is proportional to the rapidity with which the food passes through the intestinal canal. The examination of the properties of the meconium and of the intestinal contents of the fœtus generally, is a subject of more importance.

According to my experience, the small intestine of the *human fœtus*, between the fifth and sixth month, always contains a bright-yellow mass, which is either neutral or faintly acid; its ethereal extract consists of margaric and oleic acids and saponifiable fat, and when treated with sulphuric acid and sugar, only very gradually yields a purple color; in the alcoholic extract we may recognize taurocholate of soda (partly by its relations towards the salts of lead, acids, and alkalies, and partly by the formation of sulphuric acid when treated with potash and nitric acid), bile-pigment (although not always to be detected by nitric acid), and the chlorides of sodium and potassium. Boiling alcohol extracts from the mass, which is insoluble in the cold fluid, a substance which separates on cooling, and in its further reactions is similar to casein or albuminate of soda; the watery extract contains a substance precipitable only by tannic acid (unaffected by neutral or basic salts of lead or silver), and presents traces of alkaline sulphates. By far the greatest part of the solid materials in these cases (from 89 to 96% of the dry residue) consists of insoluble matter, namely, of epithelial structures and mucus.

The contents of the *large intestine* of the fœtus in and after the seventh month, are almost perfectly similar to the *meconium* discharged after birth; they constitute dark-colored, brownish-green, almost black, tolerably compact masses, devoid of odor, and without any very well-marked taste, but having a strong tendency to decomposition (as also has been observed by Höfle);<sup>1</sup> at an ordinary temperature this substance has, in the course of twenty-four hours, converted spirit of 78·8° into acetic acid. As a general rule, I have found the contents of the large intestine, as well as the meconium, acid; occasionally, however, they are neutral; under the microscope the masses are found to consist essentially of epithelium and mucus-corpuscles, the epithelium presenting a beautiful green tint; ether extracts a tolerably large quantity of fat, in which, by careful evaporation, the most beautiful tablets of cholesterin may be perceived; the alcoholic extract forms a greasy blackish-brown mass, which under the microscope exhibits no trace of crystallization; no distinct reaction either of the biliary acids (by the sulphuric acid and sugar test) or of bile-pigment (by nitric acid) could be obtained.

<sup>1</sup> Chem. u. Mikrosk. 2 Aufl. S. 85.

The watery extract, even when obtained before the substance had been treated with alcohol and ether, contains no substance which is coagulable or precipitable by acetic acid; it contains, however, a nitrogenous body precipitable by tannic acid but not by metallic salts; and it yields no trace of sulphates.

The bright yellow, semi-fluid excrements of infants at the breast contain, as was shown by Simon,<sup>1</sup> a very large amount of fat, which may naturally be referred to the milk; besides much coagulated but undigested casein; the alcoholic extract, when treated with a mixture of sulphuric and nitric acid, generally gives the well-known changes of color indicative of cholepyrrhin; and Pettenkofer's test applied to this extract usually demonstrates the presence of the biliary acids. Epithelial structures abound in these excrements.

Liebig some years ago made the remark that the solid excrements contain only a small amount of soluble salts; I found only 23·067% of soluble salts in the ash of normal human excrement; Fleitmann,<sup>2</sup> on the other hand, found 30·58% (after an abundant animal diet), and Porter<sup>3</sup> 31·58%; the latter chemist found that in dried normal excrements generally there are contained, on an average, 6·69% of mineral substances. The ash of human fæces contains, according to Fleitmann, 30·98%, and, according to Porter, 36·03% of phosphoric acid in combination with alkalies or earths, the acid being combined with three atoms of base; the former found only 1·13% of sulphuric acid, the latter 3·13%; it is singular that in the analyses of both these chemists, the potash preponderates in an extraordinary degree over the soda; if we deduct the chloride of sodium from the soluble constituents of the ash, the ratio of the soda to the potash in the ash is 1 : 40, according to Fleitmann, while it is only 1 : 12 according to Porter:—a difference which depends upon the nature of the food. Berzelius first directed attention to the fact that more lime than magnesia must be absorbed in the intestine, since we find in the solid excrements less lime and relatively more magnesia than in the food that has been taken; while the ratio of the lime to the magnesia in the fæces varies according to the nature of the food, there is always a relative excess of magnesia. In 100 parts of ash Fleitmann found 21·36 of lime with 10·67 of magnesia, and Porter 26·46 of lime with 10·54 of magnesia. Hence the ratio of the magnesia to the lime in the excrements is as 1 : 2 or  $2\frac{1}{2}$ . Alkaline chlorides occur in the excrements in very small quantity (from 1·5 to 4·4%), but carbonates are always present in the ash. Berzelius observed that sand is always mixed with the excrements, and both Fleitmann and Porter have repeatedly noticed the same fact.

The ash of the *dung of herbivorous animals* (the cow, the sheep, and the horse) has been analyzed by Rogers, and,<sup>4</sup> in essential points, is the same as that of human excrement. It contains more silica and sand, but that is easily accounted for. It is worthy of remark that Rogers found scarcely any traces of alkaline carbonates in these ashes.

*Very soluble salts* only enter into the solid excrements in large quantity, when they excite diarrhœa; Laveran and Millon<sup>5</sup> have obtained

<sup>1</sup> Med. Chem. Bd. 2, S. 488 [or Vol. 2, p. 369, of the English translation].

<sup>2</sup> Pogg. Ann. Bd. 76, S. 356.

<sup>4</sup> Ibid. Bd. 65, S. 85–99.

<sup>3</sup> Ann. d. Ch. u. Pharm. Bd. 71, S. 109–115.

<sup>5</sup> Ann. de Chim. et de Phys. T. 12, p. 135.

this result with sulphate of soda and acetate of potash, and I have done so with phosphate of soda.

The presence of crystals of *phosphate of ammonia and magnesia* in human fæces, was for a time regarded as a sign of a grave disease, namely typhus; pathologists are, however, now generally of opinion that such is by no means the case, and that these crystals often occur in perfectly normal evacuations, although it is only under specially favoring conditions that they are found in large quantity. It cannot, however, be denied that, in certain diseases of the intestinal canal, in which the secretions and the contents of the bowels are especially prone to decomposition, as in typhus, cholera, and certain forms of dysentery, these triple phosphates are found in an extraordinary quantity, on examining the evacuations by means of the microscope.

We have already pointed out that, in all cases in which the food passes more rapidly than usual through the intestinal canal, a larger quantity of *undecomposed bile* is always found; hence this is the case after the use of saline and acrid purgatives, and in the simplest forms of catarrhal diarrhoea, as Pettenkofer<sup>1</sup> himself proved. That in jaundice, dependent on occlusion of the common biliary duct, even the products of the decomposition of the bile should not occur in the stools, is a fact scarcely requiring mention. The excrements in such cases are of a dirty whitish-gray color, and develop a *very* disgusting, putrid odor; in other respects they do not essentially differ from normal fæces.

A *green coloration of the excrements* was formerly, and for a long time, regarded as a sign of the presence of bile; latterly, however, its presence in green stools has been altogether denied. The cases are certainly only few in which the green color of the fæces depends on the admixture of imperfectly metamorphosed bile-pigment, and are almost entirely limited to the condition of true polycholia, which rarely occurs in adults, but is ordinarily present in *icterus neonatorum*. In these cases the cholepyrrhin, in consequence of the predominance of free acid, appears to be converted in the intestine only into that modification of the pigment which we term biliverdin. On adding nitric acid to the alcoholic extract of these stools, we obtain the ordinary reaction of bile-pigment, and with concentrated sulphuric acid and sugar we obtain indications of the presence of the resinous acids, so that no doubt can remain regarding the abundant existence of almost unchanged bile in these stools.

Every one is acquainted with the appearance of the grass-green, pulpy stools, which so frequently *follow the administration of calomel*. There have been many experiments, but far more controversial discussions, in reference to this coloration. My own investigations lead to the following conclusions:—After calomel has been taken, we always find mercury in the stools, whether they be green, or black, or of their ordinary color; this had previously been distinctly established by Hermann,<sup>2</sup> and even more strongly by Merklein.<sup>3</sup> Höfle has likewise convinced himself of the presence of mercury in the fæces in these cases. The sulphide of

<sup>1</sup> Ann. d. Ch. u. Pharm. Bd. 53, S. 90.

<sup>2</sup> De rationibus dosium calomellæ, &c. Diss. inaug. Hauniæ, 1839.

<sup>3</sup> Ueber die grünen Stühle nach dem Gebrauche des Calomels im typhösen Fieber. Inauguralabhandlg. München. 1842.

mercury may be separated, by rinsing, from the evacuation, when stirred in water, as Merklein was the first to observe, and its chemical nature may be then very easily recognized; the dark color of the sulphide of mercury, when finely comminuted, may certainly, like sulphide of iron, give rise to a light-green color with animal substances, and especially with the yellow bile-pigment; indeed, powdered calomel, when triturated with yellowish-brown excrements, causes them, according to Hermann, to assume a greenish color. But, notwithstanding these facts, we should not deny the presence of almost unchanged bile in calomel stools, for we may with facility recognize the presence of bile-pigment by nitric acid, and of the resinous biliary acids, by Pettenkofer's test, in the alcoholic extract when carefully prepared; and this extract may usually be obtained in considerable quantity. Every one who himself analyses such stools is, at all events, led to the subjective conviction that a part of the green and light color may be dependent on bile-pigment. To this we must add that Buchheim<sup>1</sup> has recently convinced himself by experiments on dogs, provided with artificial fistulous openings (made according to Schmidt and Bidder's directions) between the gall-bladder and the external abdominal walls, that the administration of calomel actually causes an increased secretion of bile, as well as a more abundant secretion of mucus. If, moreover, the administration of calomel is sometimes not followed by green stools (and this is not very unfrequently the case), the evacuations either retaining their normal color, or presenting the characteristics of special morbid processes, this must not be regarded as presenting an argument against Merklein's view; for it is obvious that, when the intestinal canal is in an abnormal state, the conditions may not always be present which are requisite for the formation of sulphide of mercury. On the other hand, this is as little in opposition to the view that the bile-pigment takes part in the coloration, since there are various conditions under which the action of calomel on the hepatic secretion may be modified and entirely checked.

The case is altogether different with the dark, often black, but frequently also green-colored stools, which occur after the prolonged use of *preparations of iron*, or *chalybeate mineral waters*, especially such as contain sulphate of soda with carbonate of protoxide of iron. Kersten<sup>2</sup> was the first to show that the green color of these excrements was due to sulphide of iron; his only error was that he ascribed the color to the bisulphide, being led astray by the analogy with the formation of prismatic iron pyrites,  $\text{Fe S}_2$  (spear pyrites), which, as is well known, is produced in stagnating waters, when organic substances undergo putrefaction in the presence of the oxides of iron and alkaline sulphates. In three cases in which I analyzed the green and black excrements of persons who for a long time had taken the Marienbad waters at their source, I<sup>3</sup> found 3.163%, 1.039%, and 2.100% of proto-sulphide of iron in the dry residue of the pulpy stools.

The watery extract of these excrements contained much sulphate of protoxide of iron, which seemed to increase in proportion to the length

<sup>1</sup> In a private communication.

<sup>2</sup> Walther's u. Ammon's Journ. f. Chir. Th. 8, S. 180\*

<sup>3</sup> Göschen's Jahresber. Bd. 8, S. 42.



of time during which they were digested with water and exposed to the air. The residue of these excrements, which was insoluble in water, alcohol, and ether, developed sulphuretted hydrogen when treated with hydrochloric acid, and the acid filtered fluid gave distinct indications of iron with all the ordinary reagents. I now separated the residue insoluble in water, alcohol, and ether, into three parts; from one I extracted the iron with hydrochloric acid, treated the solution with chlorine, and determined the peroxide of iron quantitatively by precipitating it with caustic ammonia; the second part I treated with aqua regia, and determined the iron and sulphuric acid from the solution; while I incinerated the third part with carbonate and nitrate of soda: by these means I found that the iron stood to the sulphur in about the ratio of 28 : 16, which obviously corresponds to the protosulphide.

It has been doubted whether the sulphide of iron, even in the state of finest comminution, can give rise to a green color; but we may very easily convince ourselves on this point by adding a proto-salt of iron to albumen, dissolving the precipitate by an alkali, and passing a current of sulphuretted hydrogen through the solution, or by adding a liver of sulphur.<sup>1</sup> There is then no precipitate, but the previously colorless fluid becomes of an intense steel-green color from the sulphide of iron which is formed.

The alcoholic extract of these excrements, which was of a very faintly yellow color, contained neither bile-pigment nor the resinous biliary acids; but in the ethereal extract, there was, in addition to fat, a substance which yielded the most distinct reaction on the addition of sugar and sulphuric acid.

In the ethereal extract, which ranged from 6 to 16% of the dried excrements, there were contained not only margarin and olein, but also butyric acid, and probably some other acids of the same group. In the dry excrements there were contained from 22 to 24% of substances soluble in alcohol, from 14.5 to 18.7% of substances soluble only in water, and from 16.6 to 26.8% of insoluble matters (remains of food, mucus, &c.) The mineral substances in these excrements, after drying, ranged from 18.4 to 27.8%, of which from 3.04 to 4.67% was sulphate of soda.

Many vegetable substances likewise communicate a more or less *green* or *black color to the excrements*. The stools are often green after the medicinal use of indigo; they are often black after taking bilberries or charcoal; of a light color after the use of rhubarb, gamboge, and saffron. They are, however, also of a bright-yellow color when the bile only flows sparingly into the intestine, as in many affections of the liver.

The presence of a large quantity of *fat* in the excrements after the use of fatty food is easily accounted for, since the experiments of Bous-singault, as well as those of Bidder and Schmidt, show that only a certain quantity of fat can be resorbed in the intestinal canal: the same is observed after the use of cod-liver oil. According to Heinrich,<sup>2</sup> the amount of fat in the fæces is increased by morbid action in wasting diseases, such as pulmonary phthisis, Bright's disease, and diabetes mellitus;

<sup>1</sup> [This term includes all soluble metallic sulphides.—G. M. D.]

<sup>2</sup> Hæser's Arch. Bd. 6, S. 306.

the augmentation of fat is, however, not of constant occurrence in any of these diseases.

A solid margarin-like fat has very frequently been found in the excrements in diabetes by Simon,<sup>1</sup> Heinrich,<sup>2</sup> and others. I have, however, not succeeded in finding a decided augmentation of fat in the cases in which I have examined the excrements of diabetic patients. The loss of fat through the intestine is therefore, at all events, not a constant symptom in diabetes.

It has been asserted that *sugar* has been found in the excrements in cases of diabetes mellitus; its presence, however, is not constant.

The occurrence of *blood* in the fæces is very common, although it often escapes observation. In hemorrhoids, dysenteries, and other considerable hemorrhages of the large intestine, the presence of the blood cannot be overlooked, and, as a general rule, no manipulation or tests are requisite for its detection. If, however, the hemorrhage is very slight, and proceeds from the stomach or small intestine, the excrements appear variously colored, so that no conclusion regarding the admixture of blood can be drawn from the color and general appearance of the fæces. Every one has seen the black or chocolate-colored tar-like stools, which were formerly regarded as peculiar to melaena, but which are observed in all cases of hemorrhage in the upper part of the intestinal canal, in round (perforating) ulcer of the stomach or duodenum, in cancer, corrosions, &c. By a microscopic examination, fragments of blood-corpuscles may always be detected in such excrements, and hæmatin may be recognized chemically by means of alcohol containing sulphuric acid; in one instance (a case of cancer) I found a large admixture of colorless blood-corpuscles or mucus-corpuscles. In typhus, green fluid or semi-fluid excrements are not very unfrequently discharged when no calomel has been administered (and, conversely, it often happens that the use of calomel in this disease is not followed by the green stools which are characteristic of this medicine); and in this case the green coloration is dependent on an admixture of blood, the same as is sometimes observed in dysentery and in the intestinal diseases of young children. Bile-pigment and the biliary acids are only rarely to be detected in any quantity in such stools by a chemical investigation; if, however, we examine a portion under the microscope, we always find distorted blood-corpuscles, some distinctly yellow, and others very pale, together with colorless cells resembling pus-corpuscles. Hence it hardly admits of a doubt that, in such excrements, the green coloration essentially depends on the blood which is distributed through it; we find, however, in other secretions which are never accompanied by an effusion of blood, especially in cases of typhus, a green color, as, for instance, in the pulmonary expectoration, which, even in an ordinary case of pneumonia, very often assumes a color merging strongly on green, and in which the most beautiful blood-corpuscles may be detected by the microscope.

*Albumen* in a coagulable state sometimes occurs in normal fæces, as has been already mentioned. It is in dysentery that it is secreted in the largest quantity from the intestine; the dejections in this disease are often so rich in albumen, that, on the addition of nitric acid, or on

<sup>1</sup> Beiträge u. s. w. Bd. 1, S. 408.

<sup>2</sup> Häser's Arch. Bd. 6, S. 306.

boiling after neutralization with ammonia, the whole fluid solidifies. Coagulable albumen is also very often found in the pulpy or fluid evacuations which sometimes occur in Bright's disease. It is constantly present in tolerably large quantity in the fluid stools in typhus. In cholera, some coagulable albumen may always be detected in the evacuations from the bowels; but here, as in the investigation of most albuminous stools, we must neutralize the fluid with acetic acid before boiling, since it generally has an alkaline reaction in consequence of the presence of more or less carbonate of ammonia, or else effect the coagulation of the albumen by nitric acid, alcohol, &c. The quantity of albumen in the intestinal dejections in cholera, is, however, far less than in typhus.

*Epithelial structures* occur in the stools in all cases of diarrhœa; in typhus, cholera, and dysentery, the diarrhœa causes a rapid desquamation of the epithelium, which for the most part hangs together in masses; indeed, in cholera, we often find the entire epithelial investment of individual villi.

*Mucus- or pus-corpuscles*, are seldom entirely absent in the stools in cases of diarrhœa; they occur chiefly in simple catarrhal diarrhœa; they have sometimes been found in such quantities in the evacuations, that, from the milky appearance they communicate to the latter, the term *chylorrhœa* has been applied to this class of cases. It is in the course of chronic dysentery (lientery) that this phenomenon is most commonly observed. In typhus and in cholera we always find a great number of these cells, but they are most abundant in cases of uncomplicated dysentery.

We find a *glassy mucus* conglobated in masses of various sizes in catarrhal affections of the large intestines, both when they occur primarily, and when associated with typhus. This mucus is ejected from the follicles of the colon, and the round and pale, or elongated and granular cells and nuclei, which may be recognized in it by a microscopic examination, clearly indicate its origin.

*False membranes, fibrinous exudations, and shreds of gangrenous mucous membrane*, are found in the evacuations in typhus, croupous dysentery, and follicular ulceration.

The various *intestinal worms, hydatids, &c.*, which sometimes occur in the evacuations, do not fall within the scope of our department.

For the clearer comprehension of the subject, we shall give a condensed view of the physical and chemical relations of the intestinal dejections in certain diseases, namely, in typhus, dysentery, and cholera.

In *typhus*, the stools are usually fluid, of a yellowish-brown color (often resembling that of dry peas), of an abominable smell, and an alkaline reaction. On allowing one of these evacuations to stand for some time, there is formed a yellowish mucous sediment, in which we may observe flocculi of undigested food, white granules, and, if catarrh of the large intestine be simultaneously present, some clots of glassy mucus. The fluid has a yellowish or pale brown, turbid appearance, and contains more or less albumen. The white granules in the sediment, which are generally about the size of a pin's head, present, under the microscope, the appearance of little more than an amorphous mass, and are probably merely a product of the intestinal ulcers; the epithelium suspended in the

fluid has for the most part a yellow tinge; crystals of phosphate of ammonia and magnesia occur in the sediment in large number, and the fluid usually contains some distorted and decolorized blood-corpuscles. By means of the microscope, we very often detect *vibriones* and fungous growths of various kinds. The green color of the stools in typhus has been already noticed. The fluid lying above the sediment contains only a little biliary matter, but a very large amount of soluble salts and especially of chloride of sodium, in addition to more or less albumen.

At the commencement of *dysentery*, the intestinal discharges consist chiefly of epithelium, and of a fluid poor in albumen, and mixed with a little true faecal matter; when the process assumes a well-marked croupous character, the evacuations consist chiefly of a mixture of blood and purulent matter, in which we can detect fibrinous exudations, blood-corpuscles, cylindrical epithelium, and pus-corpuscles. When the disease runs a less severe course, clots of glassy mucus from the follicles of the colon predominate; moreover, crystals of triple phosphate may always be observed; the fluid is extremely rich in albumen, being a true exudation of the blood-plasma; biliary matters may be recognized in the alcoholic extract of its solid residue by nitric acid, as well as by Pettenkofer's test.

The stools in *Asiatic cholera* have been submitted to many analyses, which, however, have led to few results, insomuch as the simultaneous characters of the blood and of the cholera-process in general, have not been taken into consideration. The only peculiarities which we find in the stools in cholera, are the above-mentioned shreds of cylindrical epithelium, an extraordinary quantity of water, a little albumen, very little biliary matter, and a relatively large amount of salts, amongst which, according to the evidence of all observers, the chloride of sodium predominates, and often to such a degree as to exceed in amount all the organic matters. The rice-water appearance of such stools simply depends on the suspended epithelium. The rose-red tint which the fluid assumes on the addition of nitric acid would be characteristic of these stools, if the same were not also often observed in typhus. These evacuations contain only from 1·2 to 2·4g of solid constituents (Becquerel,<sup>1</sup> Güterbock,<sup>2</sup> Schmidt).<sup>3</sup>

The intimate connection of these intestinal transudations with pathologico-chemical processes in general, finds its natural place under the head of "The Metamorphoses of the Animal Tissues," and will be noticed in the second volume.

*Intestinal concretions* are rare in man and in carnivorous animals, but are comparatively common in herbivorous animals, and especially in the horse. They consist chiefly of phosphate of ammonia and magnesia, with some phosphate and carbonate of lime, which have deposited themselves around a fragment of undigested vegetable or animal food. Hence their quantitative composition presents no peculiar interest in a physiologico-chemical point of view.

The concretions termed *bezoars*, which have recently been examined with much care by Merklein and Wöhler<sup>4</sup> and by Taylor,<sup>5</sup> are of far

<sup>1</sup> Arch. gén. de Méd. 4 Sér. T. 21, p. 192.

<sup>2</sup> Journ. f. pr. Ch. Bd. 48, S. 450.

<sup>3</sup> Charakteristik der Cholera, u. s. w. S. 79, 81.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 55, S. 129-143.

<sup>5</sup> Phil. Mag. Vol. 28, pp. 44 and 192.

more importance and interest. The former analysts found that bezoars might be classified according to their chemical nature, (1) into such as consist of *phosphate of lime and phosphate of ammonia and magnesia*; (2) into such as consist of *lithofellic acid*; and (3) into those formed of *ellagic* or *bezoardic acid*. It is to the last class and its constituents that the above-named chemists have especially devoted their attention.

The *bezoars consisting of ellagic acid*, which are the true oriental bezoars, have a dark olive-green and sometimes a marbled brownish color, an oval form, a smooth surface, a concentric laminated structure, and splinter when broken; in their interior they have a foreign nucleus; their size varies from that of a bean to that of a small hen's egg. On being heated, they carbonize without fusing, and become colored with glistening yellow crystals. Like Taylor (sec p. 113), they found the bezoardic acid to be identical with the substance known as ellagic acid, but they assigned to it a somewhat different composition from that determined by Pelouze, their formula being  $\text{HO} + \text{C}_{14}\text{H}_2\text{O}_7 + 2\text{Aq}$ , while that of the French chemist was  $\text{C}_7\text{H}_2\text{O}_4$ . This acid possesses the peculiarity that in its potash-salts it oxidizes very rapidly when free access of atmospheric air is allowed, so that amongst other products of decomposition, a new acid, *glauco-melanic acid* ( $= \text{C}_{12}\text{H}_2\text{O}_6$ ), is produced. It is worthy of remark that the last-named acid, if its potash-salt be treated with water or be decomposed by hydrochloric acid, again yields ellagic acid.

The *formation of ellagic from gallic acid* during the act of digestion in animals yielding bezoars, may be explained by our assuming that two atoms of gallic acid lose three atoms of water and assimilate an atom of oxygen, as is shown in the formula,  $\text{C}_{14}\text{H}_6\text{O}_{10} - 3\text{H}_2\text{O} + \text{O} = \text{C}_{14}\text{H}_2\text{O}_7 + \text{HO}$ .

Taylor has also carefully examined the intestinal concretions known as bezoars. He divides them into (1) calculi consisting of animal hairs; (2) of vegetable hairs; (3) of ellagic acid; (4) of lithofellic acid; (5) of phosphate of ammonia and magnesia; (6) of diphosphate of magnesia; (7) of diphosphate of lime; (8) of oxalate of lime; (9) of ambergris.

Taylor describes the concretions containing ellagic acid in much the same manner as Merklein and Wöhler. These *true oriental bezoars* are not only obtained from the intestinal canal of a wild goat inhabiting the Persian province of Chorasán, but also from *Babianum cynocephalum*. When freshly obtained from the animal, they have about the softness of hard-boiled eggs.

The concretions consisting of lithofellic acid, probably originate, according to Taylor, in resinous matters taken in the food. Taylor suggests that this acid should be named *resino-bezoardic acid*.

The *excrements of birds and serpents*, which, mixed with the renal secretion, are discharged from these animals through the cloaca, as well as *Guano*, the *Hyraceum* or *Dasjepis* of *Hyrax capensis*, and the *excrements of insects*, will be fully noticed when we treat of "The Urine."

## BLOOD.

From the earliest times the blood has been made the subject of the most various hypotheses, which, however so far harmonized together, that they agreed in ascribing to this fluid the most important share in the maintenance of animal life. Moses, in accordance with the views of the ancient Egyptians, like Empedocles, placed the seat of life in the blood. This fluid therefore has in all ages played an important part in the History of Medicine. One might therefore reasonably have expected that the inquirers of modern times would have been in possession of more than sufficient empirical supports on which to establish with some degree of completeness a knowledge of this most subtle of all animal fluids; but unfortunately the methods accessible to earlier investigators were so imperfect and their modes of inquiry so widely different from those of the present day, that even the discoveries of the last century have been of little service in enlarging our views on this subject. We need hardly allude to the obstacles opposed by a mere transcendental philosophy based upon vague notions of vitality and vital forces, by a deficient knowledge of physics and even of logic, for when we call to mind, that only three-quarters of a century ago oxygen was unknown to the chemist, we have at once a ready explanation of the inability which formerly existed of elucidating the great mysteries of animated nature. Even physics, which had solved some of the great problems of astronomy, were still incapable of interpreting the phenomena of the animal organism. It is only from a comparatively recent period that we can date the first moderately accurate microscopical investigations of the blood-corpuscles, and the first attempts to investigate their origin, function and destiny, or an accurate and systematic mode of analyzing the blood, &c. In the present day the investigation of the blood has been conducted with the most earnest attention and the most zealous activity; yet notwithstanding the devotion of so much labor, the theory of the blood is yet in the first stage of its development. But it must be remembered that the mass of correctly or incorrectly observed facts and of more or less ingenious hypotheses is abundant in proportion to the recent date of a science and to its want of fixed and reliable points of support. Such has been the fate of the theory of the blood. Its right comprehension has been rendered nearly impracticable amidst the accumulation of the innumerable inquiries which have been instituted in reference to its physical and chemical relation in physiological and pathological conditions, and amidst the multifarious and contradictory views promulgated regarding its progressive and regressive metamorphosis and the functions of its various constituents individually and collectively; so that it is now alike impossible to afford a clear and succinct exposition of the theory of the blood, and to sift facts from conclusions, or the positive from the merely hypothetical. In a chemical point of view this somewhat unpromising prospect must be referred to an imperfect knowledge of the true basis of the whole inquiry; and all who have attentively followed our observations on the protein-compounds, the mineral substances of the animal body, the pigments, &c., will clearly comprehend that no tho-

rough knowledge of the blood can ever be obtained until we shall succeed in throwing some degree of light upon these obscure departments of zoo-chemistry.

The blood, as it flows in the vessels of the higher animals, is a somewhat tenacious fluid, heavier than water, and presents various shades of red; in the arteries, however, it is constantly somewhat brighter than in the veins; it is only transparent in very thin layers. Immediately after its removal from the circulation, it becomes more tenacious and gelatinous, and finally separates into a firm, dense red mass, and a clear faintly yellow fluid.

From accurate inquiries regarding the physical properties of the blood, it has been ascertained that the *specific gravity* of normal human blood averages 1.055, its physiological limits being 1.045 and 1.075; in women it is rather less than in men, and in children than in adults; in pregnant women it is even lower than in women who are not pregnant.

Nasse,<sup>1</sup> whose labors have contributed very much to our knowledge of the blood, found that its *capacity for heat* stood in an exact ratio to its density.

The *color* of the blood, as we usually see it, may be described as a bright cherry-red; it is clearer in early youth than in the foetal state, in infancy, or in old age; it is somewhat darker in pregnant than in non-pregnant women. The use of various kinds of food and drink, bodily exercise, and other physiological relations, to a certain degree influence the depth of the blood's color. The action of gases and other substances on the color of the blood will be noticed in a future page.

While still warm, the blood has a peculiar *odor*, which is generally somewhat stronger in men than in women.

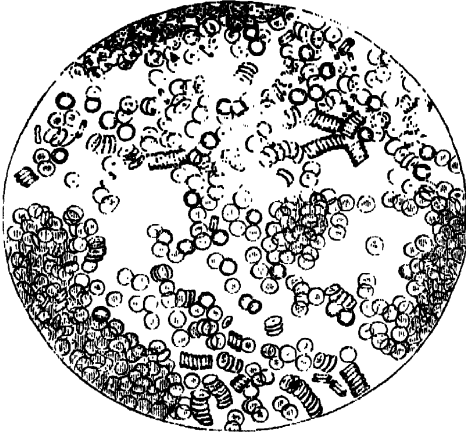
The blood *coagulates*, the process commencing in from two to five minutes after its abstraction, at the surface and edges, when it gradually becomes tough and gelatinous; in the course of from seven to fourteen minutes, the jelly which is thus formed has attained such a consistence that the whole mass has assumed the form of the interior of the vessel, and has lost all its fluidity. The separated substance through which the whole blood has been converted into a jelly, now begins gradually to contract, so that a great part of the fluid enclosed by it is pressed out towards the surface; this expressed fluid we name *serum*. The contraction of the gelatinizing substance continues for a period varying from twelve to forty hours, during which time a dense red clot or coagulum, the *blood-clot*, is formed; this usually assumes the form of the interior of the vessel on a reduced scale; the lower part of the clot is generally darker, and the upper of a brighter red than the original uncoagulated blood. In men the coagulation proceeds more slowly, but the coagulum is denser than in women. Arterial blood coagulates more rapidly than venous. Atmospheric air hastens the coagulation; regarding the influence of temperature on this process, there are still different opinions. By shaking, stirring, or whipping freshly-drawn blood, the coagulating substance separates in yellowish flocculi or pellets, while the fluid remains as red as the uncoagulated blood (or perhaps a little lighter in tint) and equally untransparent.

<sup>1</sup> Handwörterbuch der Physiol. Bd. 1, S. 79.

Since the times of Malpighi and Leeuwenhoek, it has been known that the blood is not a simple solution of various substances, but an emulsive fluid in which solid particles are contained in suspension. These solid particles consist principally of the blood-corpuscles, with which, however, other formal elements are mixed, although in far less quantity. Recent *microscopic* observations have revealed to us the following facts in relation to the blood-corpuscles.

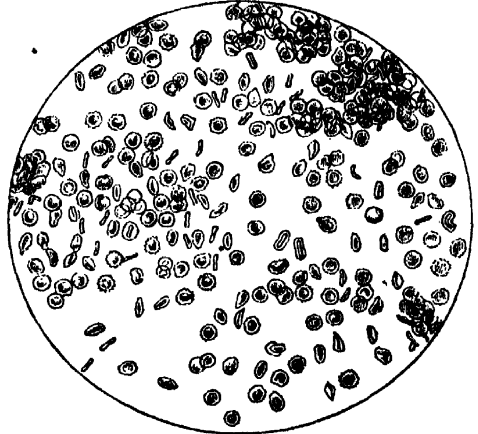
The *blood-corpuscles* or *blood-globules* are distinguished by peculiarities of form and size in every animal genus: in man they are thick, circular, slightly biconcave disks consisting of a colorless investing membrane, and red, or, in refracted light, yellow, viscid fluid contents. Most observers

Fig. 21.



Coagulation of normal human blood under the microscope.

Fig. 22.



Human blood-corpuscles, treated with a concentrated solution of sulphate of soda.

at present coincide in believing that these corpuscles have, except in rare instances, no true nucleus, a very few occasionally containing an indistinct, light granule in the concave centre. The blood-corpuscles of other mammalia likewise form round disks, except those of the camel, the dromedary, and the llama, which are elliptic and biconvex. In birds the corpuscles are elongated and oval, elevated in the centre, and have a sharply defined outline; in amphibia they are oval and strongly convex.

The human blood-corpuscles average about one three-hundredth of a Paris line ( $=0.00333'''$  or  $0.00752^{mm}$ ) in diameter. E. H. Weber and R. Wagner have shown that in the embryo these corpuscles are generally somewhat larger than in the same animal after respiration has been established. The blood-corpuscles of the mammalia approximate tolerably closely in size to those of man; they are, however, all somewhat smaller: those of the other vertebrata, and especially of the amphibia, are, however, far larger (ranging to  $0.0142'''$  or  $\frac{1}{72}'''$ ); the largest occur in the blood of *Proteus auguinus*.

The difference in size of the blood-corpuscles of different animals, is a point of the greatest importance in the investigation of their blood, and is the more deserving of attention, since here, as in so many other cases, chemistry entirely fails us, while the microscope and micrometry yield



the most decisive results. It is at present utterly impossible to tell with certainty by chemical analysis, to what species of animal a specimen of blood that may be presented to us for examination belongs. For this object various means have been devised, to which we shall, in a future page, at all events, make a passing reference: but none of them yield such decisive results in the majority of cases as the microscopico-mechanical analysis. From what has been already stated, it is obvious that by the microscope, and independently of all other aid, we can readily distinguish the blood of the different classes of vertebrata. C. Schmidt<sup>1</sup> has, however, shown that by accurate microscopical measurements of the corpuscles, the blood of many of the mammalia can be individually detected, and distinguished from that of man. The differences in the size of the corpuscles of different animals are so very small, that the ordinary methods of measuring these minute bodies by Weber's glass micrometer or a screw micrometer, are by no means sufficient. The blood-corpuscles being vesicles or cells with an extremely thin investing membrane, are very much influenced by endosmotic currents, and it is clear that their diameters must vary with the quantity of fluid they have imbibed or given off. When the serum loses water by evaporation, a current of fluid passes out from the corpuscles to the serum; their diameter must then diminish, just as on the addition of water we see it increase. As in the ordinary methods of measuring the blood-corpuscles, gradual evaporation cannot be prevented, and the coefficient of evaporation for each special case cannot be calculated, the idea suggested itself to Schmidt of measuring the corpuscles of *dried* blood. On drying fresh blood in extremely thin layers on a glass-plate, the corpuscles lie with their flat surfaces on the glass, adhere to it, and remain extended on it after drying. Schmidt has carefully ascertained by numerous measurements that the mean diameters of the blood-corpuscles in this dried and extended membrane-like state, are unaffected or only slightly diminished, that at least from 95 to 98% of the corpuscles of the blood of the same animal species are of equal size, and finally, that the corpuscles of the blood of different species of the mammalia present constant differences of size. The following are, according to Schmidt, the diameters of the blood-corpuscles in this state, expressed in millimetres:

	Mean.	Minimum.	Maximum.
Man, . . . . .	0.0077	0.0074	0.0080
Dog, . . . . .	0.0070	0.0066	0.0074
Rabbit, . . . . .	0.0064	0.0060	0.0070
Rat, . . . . .	0.0064	0.0060	0.0068
Pig, . . . . .	0.0062	0.0060	0.0065
Mouse, . . . . .	0.0061	0.0058	0.0065
Ox, . . . . .	0.0058	0.0054	0.0062
Cat, . . . . .	0.0056	0.0053	0.0060
Horse, . . . . .	0.0057	0.0053	0.0060
Sheep, . . . . .	0.0045	0.0040	0.0048

These investigations constitute the first step to the diagnosis of the blood of different kinds of animals.

*Colorless blood-corpuscles*, or as they have been termed, lymph-corpuscles, are constantly present in the blood, although in far less number than the colored corpuscles: they are more globular, although not per-

<sup>1</sup> Die Diagnostik verdächtiger Flecke in Criminalfällen. Mitau and Leipzig, 1848.

fectly spherical, and their diameter averages  $\frac{1}{200}$ ''' (which is 0.005''' or 0.0128<sup>mm</sup>); they have a granular capsule, and either a single round, or occasionally an oval or reniform nucleus, or several small nuclei heaped on one another; in consequence of their containing a larger quantity of fat, and of their being deficient in the ferruginous hæmatin, they are specifically lighter than the red corpuscles: these cells were formerly regarded by some writers as products of coagulation, but independently of their appearance under the microscope, which shows that they unquestionably are cells, we may also readily convince ourselves that they are contained preformed in the living blood by a microscopical examination of the circulation in the capillaries of the frog (in the web-membrane, the mesentery, or the tongue).

It is seldom, except in whipped blood, that other formal elements, namely, fat-globules and the so-called fibrinous flakes (*Faserstoffschollen*), are detected by the microscope.

The fluid in which the blood-corpuscles are suspended, has received the names of *Liquor sanguinis*, *Plasma*, and *Intercellular fluid*; in the circulating blood it contains in solution, together with other matters, the substance on which the coagulation of the blood depends.

The *Clot* consists of the coagulable matter of the blood together with the blood-corpuscles, which it encloses in the process of separation; it is moistened by a varying amount of serum.

The *specific gravity* of the serum is less variable than that of the blood itself; it averages 1.028.

The blood, consequently, is divisible mechanically into three parts: the coagulating substance, the serum, and the blood-corpuscles. Hence, nature appears to aid the efforts of the chemist, who, next to a perfect chemical separation, always desires also to accomplish a mechanical separation of the constituents of the object which he is engaged in examining; but unfortunately this very circumstance constitutes one of the principal grounds which have hitherto stood in the way of a scientifically accurate analysis of the blood. The impracticability of a perfect separation of the blood-corpuscles, which are moist cells filled with water and soluble substances, from the surrounding intercellular juice (which we shall presently examine), always prevents the possibility of our obtaining any certain approach to the knowledge of the processes which are at work in the blood, and which principally consist in the reciprocal action of the cells contained in it, and the plasma surrounding them. In every investigation it is necessary—and in the consideration of this most important of all the fluids, it is especially the case—that we should, before all things, make it perfectly clear from what point the inquiry should be undertaken. The physiologist is aware that in the blood, the cells and the intercellular fluid uninterruptedly act upon one another, without however the reactions being perfectly equalized; we know that the intercellular fluid acts on the cells, and that the contents of the latter react on the former; in a word, we know that the contents of the cells and the intercellular fluid are different and heterogeneous; for if there were not a difference between them, no reaction would be conceivable.

Hence, like the fluid in which yeast-cells are developed, and indeed

like all germ-fluids, the intercellular fluid contains the material from which the cells are formed, as well as the substances which are produced by the activity of the cells or their metamorphosis and regressive formation.

This is the point of view from which the physiologist would desire that the investigation of the blood should commence; for thus only could fruitful results be expected. The chemist, however, whether designedly or undesignedly, has failed in sufficiently separating these different objects of investigation, and for the most part has treated them simply as different constituents of one and the same fluid, whilst he has regarded the most important morphological elements,—the most essential factors in the metamorphoses of the blood—merely as simple constituents of the collective mixture, and has placed them in the same chemical category with fibrin and albumen, after separating them, like the latter, from the particles with which he presumes they are only mechanically connected and intermingled. Such a chemical mode of treating the blood must be very detrimental to physiological progress, for the chemist is hardly able to obtain in a perfectly isolated state, the substance which he regards and calculates as dried corpuscles. Hence, in order not to be led astray in our view of the composition of this animal juice by the chemical duties of analyzing the blood and of ascertaining its constituents—points which have been already often noticed—we shall specially consider the intercellular fluid and its constituents on the one side, and the blood-cell and its contents on the other, each independently of the other. Hence, in order to make the comparison as plain as possible, we shall give in parallel columns the quantitative relations of these two great factors in the composition of the blood.

1000 parts of Blood-corpuscles contain:		1000 parts of Liquor-sanguinis contain:	
Water, . . . . .	688.00	Water, . . . . .	902.90
Solid constituents, . . . . .	312.00	Solid constituents, . . . . .	97.10
<hr/> Specific gravity, . . . . .		<hr/> 1.028	
Hæmatin, . . . . .	16.75	Fibrin, . . . . .	4.05
Globulin and cell-membrane, . . . . .	282.22	Albumen, . . . . .	78.84
Fat, . . . . .	2.31	Fat, . . . . .	1.72
Extractive matters, . . . . .	2.60	Extractive matters, . . . . .	3.94
Mineral substances (without iron), . . . . .	8.12	Mineral substances, . . . . .	8.55
<hr/>		<hr/>	
Chlorine, . . . . .	1.686	Chlorine, . . . . .	3.644
Sulphuric acid, . . . . .	0.066	Sulphuric acid, . . . . .	0.115
Phosphoric acid, . . . . .	1.184	Phosphoric acid, . . . . .	0.191
Potassium, . . . . .	3.328	Potassium, . . . . .	0.323
Sodium, . . . . .	1.052	Sodium, . . . . .	3.341
Oxygen, . . . . .	0.667	Oxygen, . . . . .	0.403
Phosphate of lime, . . . . .	0.114	Phosphate of lime, . . . . .	0.811
Phosphate of magnesia, . . . . .	0.078	Phosphate of magnesia, . . . . .	0.222

In this representation of the quantitative relations of the constituents of the principal elements of the blood, we have preferred following the experiments and deductions laid down by Schmidt<sup>1</sup> in his admirable

<sup>1</sup> Charakteristik der Cholera, u. s. w.

essay; determining from our own analyses only the mean numbers for the individual constituents, and extending our comparative tables to the fat. In describing the analysis of the blood, we shall enter fully into the methods by which those numbers, which we regard as approximative determinations, have been obtained.

From what has been already stated, it follows that in our consideration of the blood we must, in the first place, regard the morphological elements, and especially the cells, as being altogether distinct from the intercellular fluid. We shall commence, then, with the blood-corpuscles. If we would not rest satisfied with the exploded conjecture that the blood-corpuscles are living beings, whose properties are dependent on a peculiar vital force, but would attempt to form a truly logical and distinct idea of them, we must endeavor to establish an intimate relation and an organic connection between the individual phenomena which we observe in them. In accordance with the main idea that the blood-corpuscles are vesicles filled with a dark brownish-red, tenacious fluid, their individual properties must be grouped together in the most intimate relation to one another, like the edges and angles of a crystal to its axes. Thus the color of the red molecules of the blood is no incidental property, but, to its most delicate modifications, it results from the idea of a vesicle which is filled with red fluid; the forms and dimensions of these vesicles are essentially changed by various endosmotic influences, and thus give rise to the various shades of color. The form, the tendency to sink, and the specific gravity, are also properties which always have a definite relation and connection. If, therefore, we consider the physical properties of the blood-corpuscles from this point of view, we shall attain the clearest conception of their nature, and obtain the firmest basis for the support of our views regarding the mechanical metamorphosis of matter occurring between these cells and the fluid surrounding them.

One of the properties which we observe, both in whipped—*i. e.*, defibrinated blood—and also in blood on which that operation has not been performed, is the tendency of the colored molecules of the blood to sink, to a greater or less extent, in the intercellular fluid. The difference in this *sinking tendency* of the corpuscles is often extremely striking, both in physiological and pathological conditions; this phenomenon must consequently depend on some other properties of the blood-cells. This difference was formerly ascribed to the greater or lesser specific gravity of the blood-corpuscles. It was generally believed, in accordance with the views of Nasse, that this hypothesis was confirmed by the constantly-observed fact that the colorless blood-cells did not participate in this property with the red corpuscles, which differ so essentially from every other cell-formation in the character of their contents, which are of a tenacious fluid nature, and abound in iron. To determine the value of this hypothesis by a more accurate investigation, it would be necessary to institute more accurate and comparative measurements of the density of the blood-corpuscles and of the plasma; but, unfortunately, such determinations were not so easy of attainment to the earlier investigators as they have since become through the ingenious deductions and investigations of C. Schmidt. It was, however, obvious, without

such accurate measurements, that at all events the specific gravity could not be the sole cause of this sinking tendency; for it was long known that, by the addition of certain substances to the blood, the sinking of the colored cells was accelerated, while it would naturally be expected that the intercellular fluid would have been rendered denser by holding these substances in solution, and that the assumed differences in the densities of the cells and the surrounding fluid would have been more equalized. We should *à priori* have expected the very opposite—namely, a diminished sinking tendency.

Before, however, we proceed to notice the further causes of this phenomenon, let us more closely consider the *specific gravity* of the blood-corpuscles in its relation to that of the plasma. Since we cannot so completely isolate the blood-corpuscles from the plasma as to determine their specific gravity in a direct manner, it is only by an indirect method—that is to say, by a calculation based on other determinations—that we can ascertain their density in the condition in which they exist in fresh blood. Moreover, it will be shown by a subsequent analysis of their proximate constituents, that the blood-corpuscles of different specimens of blood must have a variable specific gravity; but it might even *à priori* be inferred that their density must vary with the varying constitution of the surrounding fluid, since a continuous diffusion-current exists between the contents of the cell and the intercellular fluid. Hence the density of the blood-corpuscles will not merely vary according to the quantity of ferruginous hæmatin which they may contain, but also according to their absorption or loss, according as they are in solutions more or less concentrated than their own contents; indeed, we shall presently see that the density of the blood-cells is far more dependent on the substances which are taken up or given off by endosmosis, than on the quantity of hæmatin they may contain; for this latter is far less variable than the quantity of water, and is also partially compensated for by an augmentation or diminution of fat in the blood-cells. The blood-corpuscles of healthy human blood have a density which, in man, varies from 1·0885 to 1·0889, and in woman, from 1·0880 to 1·0886. In diseases, the density is not confined within these limits. Thus, in cholera, Schmidt found that the specific gravity of the blood-cells was increased to 1·1025 or even to 1·1027, while in dysentery it was diminished to 1·0855, in albuminuria to 1·0845, and in dropsies to 1·0819.

We are indebted to the intelligence and indefatigable perseverance of C. Schmidt<sup>1</sup> for our knowledge of the density of the blood-corpuscles, as well as for many other discoveries in relation to the blood; when, in accordance with the method presently to be given, we have determined the weight of the moist corpuscles occurring in a specimen of blood, their density may be easily found by a simple equation, as soon as the specific gravities of the serum and of the defibrinated blood are known. If we assume, for instance, that a specimen of blood contains 496 p.m. of moist blood-cells, besides 4 p.m. of fibrin, that the specific gravity of the serum is 1·0280, and that of the defibrinated blood 1·0574, then we may very readily determine the density of the blood-cells by the following considerations:

<sup>1</sup> Op. cit.

996 parts of defibrinated blood occupy the space of	941·93 parts of water.
500 parts of serum	486·38

hence 496 parts of blood-cells	“	“	455·55 parts of water,
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and, consequently, the density of the blood-cells in this specimen of blood must be 1·0888.

We now revert to the sinking tendency of the blood-corpuscles, and its causes. If we microscopically examine a specimen of blood in which the corpuscles sink with extreme rapidity, we find, as a general rule, that the blood-disks lie with their sides in contact with those of the adjacent disks, and thus form masses resembling rolls of money (the nummular arrangement); while in blood in which the serum and clot only separate slowly, the corpuscles for the most part appear isolated. If from this it would appear, that the nummular aggregation or cohesion of the blood-corpuscles is the proximate cause of the more rapid sinking, the more remote cause must be sought in a greater viscosity of the parts in question. Henle believed that this property was especially dependent on the tenacity of the intercellular fluid, and on the viscosity of the cells that was thus induced: in accordance with a similar view, many had previously regarded a superabundance of fibrin in the blood as the cause of the cohesion of the corpuscles; but independently of the circumstance that numerous observations (made with the view of deciding the question) prove that there is no connection whatever between the rapidity with which the corpuscles sink and the proportion of fibrin in the blood, the perfect inertness of the fibrin in relation to this phenomenon is indicated by the fact that the corpuscles sink just as rapidly or just as slowly in defibrinated blood as in blood which contains its fibrin.

Hence the fibrin, at all events, exerts no influence on this phenomenon. There then seemed to be a tendency to ascribe the cohesion of the corpuscles to a great excess of albumen. In favor of this hypothesis the following facts were adduced, namely, that the addition of albumen or of other viscid solutions, as, for instance, of sugar and gum, hastens the sinking of the blood-corpuscles, and that horses' blood, in which the cells sink with unparalleled rapidity, contains an especially viscid and tenacious serum. Whatever probability this view might at first sight appear to possess, it will not bear a closer scrutiny: inflammatory blood, in which we most frequently observe an increased rapidity in the sinking of the red corpuscles, never contains an excess of albumen, but, on the contrary, generally contains less than normal blood; solutions of sugar and gum hasten the sinking of the corpuscles, but deprive them of their property of cohering; and lastly, the corpuscles of horses' blood sink in the serum of human or other animal blood with almost the same rapidity as in their own serum; whilst the corpuscles of other animals, when placed in the serum of horses' blood, do not by any means exhibit a great tendency to sink. Generally speaking, physical considerations do not appear to support this view. For if the viscosity of a fluid depends on the amount of attraction which its molecules exhibit towards each other, the cohesion of the particles of fluid must overcome the adhesion to the cell-walls; hence no cohesion of the blood-cells can be directly dependent on the viscosity of the fluid; but if the viscosity of the fluid

consists in the fact that its molecules exhibit a greater attraction to the cell-walls than to one another, every cell must be surrounded by a sphere of fluid by which its closer contact with other cells (and consequently the aggregation of the cells generally) is prevented; moreover, we find that in emulsions the aggregation of the suspended molecules diminishes in proportion to the tenacity and viscosity of the emulsive solution. Hence Nasse was led to seek the cause of this aggregation, not in the fluid, but in the corpuscles themselves—that is to say, in a viscid property of their capsules; he referred especially to the action of carbonic acid. An abundance of carbonic acid in the blood (whether caused by an imperfect interchange of gases in the lungs, or artificially introduced into it), is certainly usually accompanied by a rapid sinking of the blood-corpuscles. But that the membrane of the blood-corpuscle, or that its contents should actually be rendered more viscid by carbonic acid, would not be inferred from the converse experiment that oxygen and salts communicated to the blood-corpuscles a clearly deflated, smooth, although often folded surface; for a solution of sugar acts on the form of the corpuscles, as far as the change can be followed by the microscope, in just the same manner as salts, and yet induces a rapid sinking of the red blood-cells. Moreover, it is hardly probable that carbonic acid should render the blood-cells viscid, and tend to make them assume the nummular arrangement, since we may very readily observe in fresh blood the commencement and gradual formation of these rolls of corpuscles under the microscope; in the minute drops which we use for microscopical observation any excess of carbonic acid would disappear in the process of manipulation, and in every case more oxygen would be taken up than is contained in fresh blood. Hence each of these three different modes of explaining the tendency of the blood-corpuscles to sink is opposed by definite facts which at present do not allow of any satisfactory explanation of the phenomenon. Only this much appears to be distinctly established, that, in addition to the influence exerted by the relative density of the corpuscles and the serum, their viscosity must essentially promote their aggregation. Moreover, we never observe this cohesion or peculiar aggregation of the red cells in the blood while still circulating. The tendency of the red corpuscles to sink is usually very distinctly observed in the blood of persons with inflammatory diseases, or in such blood as contains a diminution of the salts and a relative increase of the albumen. A great sinking tendency of the blood-cells is very often accompanied by a watery *liquor sanguinis*. When the blood-corpuscles are dark-colored (and may therefore be regarded as rich in hæmatin or iron), they have a tendency to sink very rapidly, and to form nummular rolls; when they are of a pale color (and are rich in fat), they only sink slowly. The blood-corpuscles of the horse, which sink more rapidly than those of any other animal, are comparatively poor in fat. Repeated venesections increase the tendency of the blood-cells to sink; they then become richer in hæmatin, as has been shown by C. Schmidt; they have thus become relatively heavier, and therefore sink more readily; the increase of hæmatin in the corpuscles is here certainly only relative; in consequence of the diluted plasma, an excess of globulin is abstracted from the blood-cells, which thus become comparatively richer in hæmatin, and poorer in globulin. If we con-

sider these facts—and we shall presently notice some additional ones—we certainly feel inclined to ascribe a greater influence than we formerly did to the difference of the densities of the blood-cells and the intercellular fluid, in relation to the sinking of the corpuscles.

In special cases several conditions are often simultaneously present, which exert an accelerating or impeding influence on the sinking of the blood-corpuscles; thus, for instance, the colored cells of the blood of the hepatic veins of the horse sink very little, while those of the blood of the portal vein (simultaneously collected from the same animal) possess a very considerable sinking tendency; this may be very readily explained by the preceding observations; the difference between the density of the cells and of the serum is far more considerable in portal blood than in that of the hepatic veins; in the former, according to my experiments, the density of the serum is to that of the cells as 1 : 1·062, and in the latter as 1 : 1·053. The serum of the blood from the hepatic veins contains (relatively to the other constituents) far less albumen than that of the portal vein; and, lastly, the corpuscles of the former blood are far poorer in hæmatin than those of the latter.

It is moreover not impossible that, at all events in certain cases, there is a converse relation of the causal action to that which we have hitherto assumed; for it is quite conceivable that the sinking of the cells is less dependent on their adhesion, than the adhesion on the gravity of the corpuscles: the viscosity of the cells can at all events not be great; for the slightest mechanical actions are sufficient to break up the nummular rolls and their branches into fragments consisting only of a few cells, which, if there were any considerable degree of viscosity, would be impossible. We may regard the following as the order in which the phenomena occur: a more or less active motion is excited in the molecules of the blood, by the difference in the gravity of the blood-cells and the intercellular fluid; and the more active the motion is, so much the more frequently will the cells be pressed together, and have the opportunity of cohering, in the same manner that a fresh precipitate, as, for instance, of chloride of silver, conglomerates much more readily when the fluid and the precipitate are well stirred. If an approximation of the blood-corpuscles is rendered possible by the motion excited in the above mentioned manner, these discoid bodies can scarcely attract and adhere to each other, except by their surfaces; and that the plasma subsequently would offer less resistance to the sinking of the rolls than to that of the individual corpuscles, might be inferred from the analogous case of the chloride of silver, even if it were not obvious from physical laws. Nothing, however, but an extensive series of comparisons between the density of the blood-cells and that of the plasma, and a combination of these results with the observed sinking tendency, can enable us to come to a certain decision regarding this view: at present we must, at all events, assign to the density a considerable part of the sinking tendency of the blood-cells.

According to Nasse, the tendency of the blood-corpuscles of different animals to sink, decreases in the following order: the horse, the cat, the dog, the rabbit, the goat, the sheep, the ox, birds, the pig; so that in the horse the corpuscles sink the most rapidly, and in the pig (at all events in the winter) the most slowly.



As the density and form of the blood-cells stand in definite relations to their sinking tendency, so also is there a certain dependence between the *color* of the blood-corpuscles and their form.

It has been already shown that the coloring matter of the blood exists only in the cells, and that consequently the color of the blood is primarily dependent on the blood-cells. In reference to the color of the individual blood-cells, we always remark, on a careful microscopic examination, some which are paler and darker than the rest, although the number of those presenting an intermediate tint very greatly preponderates; in the blood of the portal vein, we always find some which have a speckled appearance, showing that the pigment is not distributed uniformly in them, as in all other blood-corpuscles. This difference, therefore, depends upon the absolute amount of hæmatin which they contain; but the color of the cells must be relatively pale or intense, according as they are dilated or collapsed by the absorption or the loss of water. The gases, especially oxygen, probably exert a chemical action on the pigment, and thus influence the coloration of the corpuscles. The color of the individual cells has, however, only a secondary influence on the coloration of the mass of the blood, but the peculiar tint of the blood is especially modified by their number as well as their form. It need scarcely be remarked, that blood which is poor in corpuscles is of a bright red color, while blood which is rich in them must be of a darker color; but notwithstanding, it by no means happens (as is shown by the beautiful investigations of Popp) that blood which is poor in cells, should invariably be pale, and that blood abounding in them should be dark-colored. Hence there must exist yet other causes which exert an essential influence on the color of the mass of the blood—causes even more important than the color and number of the cells. We are indebted to the genius of a Henle, for the first indication of a connection between the color of the mass of the blood and the form of the red corpuscles. We had been previously satisfied with the idea, that everything relating to the color of the blood pertained to chemistry, which however could yield no information on the point. The striking changes which are induced in the color of the blood by (chemically speaking) very indifferent substances, as, for instance, sugar and neutral alkaline salts, soon led observers to support Henle's view, and amongst them we must name one of the first of our hæmatologists, H. Nasse. If we dilute blood with water, it assumes a dark-red color; if the blood were previously dark-colored, it becomes still darker on the addition of water; if, in these cases, we examine the blood-corpuscles under the microscope, we find them distended, and observe that they have almost lost their discoid form and become spherical; the blood collectively must therefore appear darker, since each individual corpuscle has become converted into a spherical mirror, from which the red rays are scattered and reflected. We observe the reverse on treating the blood with neutral salts, syrup, or in short, any such substances as render the intercellular fluid relatively denser: a diffusion-current is established from the cells towards the intercellular fluid, in consequence of which, the former must collapse. A microscopic examination shows us that the collapse of the corpuscles by exosmosis causes the central depression to become more considerable, and the indi-

vidual cells to resemble concave mirrors. It is believed that the lighter color of such blood must be referred to the reflection of the red rays.

Scherer,<sup>1</sup> who has very carefully studied this influence of the form of the cells on the color of the blood, indicates also another physical cause, which exerts an influence on its coloration. The change in the form of the cells must be accompanied by a thickening or an attenuation of the investing membrane. It is obvious that when the capsule becomes thinner by the expansion of the blood-corpuscles, the pigment must shine through more in its natural, that is to say, its dark red color, and consequently, must impart a dark coloration to the mass of the blood; and that when the corpuscles are diminished, their capsules must become thickened or thrown into folds, and must thus to a certain degree conceal the true color of the hæmatin. In a somewhat similar manner, Mulder believes that the reason why arterial appears of a lighter red color than venous blood, is, that the corpuscles of the former are surrounded by a dense layer of binoxide of protein, while those of the latter possess a thinner investing membrane. Hence Mulder agrees with von Baumhauer in the belief that alkalies and dilute mineral acids communicate a dark color to the blood since they swell up the investing membrane, which is rich in binoxide of protein, and thus render it more transparent; the dark color of the blood of the portal vein is therefore owing to the quantity of alkali contained in it.

Notwithstanding the praiseworthy investigations which have been carried on in reference to this subject, and have elicited many facts confirmatory of the above-mentioned views, the study of the changes which the color of the blood undergoes, in consequence of alterations in the form of the cells, seems still to demand a more searching inquiry. Harless has already made a very promising beginning in reference to this subject. He has examined the influence of gases on the blood-cells, and although he merely experimented on the large, elliptical, biconvex corpuscles of the frog, he has not only confirmed several former observations, but has likewise thrown unexpected light on several points in connection with this question. Thus for instance, we formerly ascribed to oxygen solely a chemical part in its action on the color of the blood, although it was known that it could be removed from the blood in a mechanical way, namely, by diffusion in other gases, or by the air-pump; but Nasse, Scherer, and Harless have obtained actual proofs of the accuracy of Henle's assumption, that both oxygen and carbonic acid give rise to changes in the form of the blood-corpuscles, on which the brighter or the darker redness of the mass of the blood depends.

Although Müller did not expect that oxygen and carbonic acid would exert any visible action on the form of the blood-corpuscles, H. Nasse asserted that he had found, from often repeated experiments, that the discoid corpuscles of the mammalia become more opaque in their centre by carbonic acid, that the outer edge becomes broader, and that thus the whole vesicle swells, while after the action of oxygen the central depressions of the cells, as well as their outlines, become more distinct; and this statement is completely borne out by the observations made by Harless on the blood-corpuscles of frogs; after the action of oxygen on frogs'

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 1, S. 288.

blood he found the long diameter of the corpuscles =  $0.011'''$ , the transverse diameter =  $0.009'''$ , their form strongly elliptical, their outlines dark, the cell-wall very finely granular, the nucleus of a roundish oval form but not very distinct, and the contents of a pale yellow color; while after the application of carbonic acid, the long diameter was increased to  $0.014'''$ , and the transverse diameter to  $0.007'''$ , the form was almost spherical, the capsule as clear as glass, the nucleus distinct and with a sharp outline, and the contents redder than in the previous experiment.

The simultaneous action of the neutral alkaline salts, and of several other chemically indifferent bodies, on the form of the corpuscles and the color of the blood, has certainly been already carefully examined from various points of view, but notwithstanding this, the subject still requires a systematic investigation, in order to establish definite relations between the form of the blood-cells and the color of the blood in connection with the amount of concentration of the solutions, the temperature, and other external conditions. For the changes which the forms of the blood-corpuscles undergo are not limited merely, according to the laws of diffusion, to a simple spherical expansion, or to a flattening and a deeper central depression, but in blood obtained during disease we very often find flatly-pressed, jagged, indented and granular, or altogether distorted yellow corpuscles, and we also observe similar modifications induced by the artificial addition of various concentrated solutions of chemically indifferent substances. At present, not even an ideal connection has been established regarding the influence which the form of these jagged, star-shaped corpuscles or the nummular rolls exert on the color of the blood; indeed objectively the color co-existing with such forms has not been sufficiently observed. In reality this only is established, that all substances which dissolve or in any way destroy the investing membrane of the blood-corpuscles, or which cause it to burst, so that their contents become mixed with the intercellular fluid, communicate an intensely dark brownish-red or almost black color to the blood; while, on the other hand, all those which cause a shrivelling of the cells or a folding or thickening of the investing membrane, give to the blood a lighter red color, indeed during the first moments of their action almost a vermilion tint.

Henle was correct in his assertion, that in fresh blood, even when there is no disease, we observe other forms than those usually presented by the blood-corpuscles, and that in some specimens of blood the corpuscles more readily assume a jagged form than in others. This alteration of form is therefore only a consequence of influences which act on the blood submitted to examination; the predisposition to this change of form varies, however, in different blood, just as the urine in various acute diseases may be earlier or later in assuming its acid character and in depositing crystals of uric acid. All that we know regarding the manner in which the blood-corpuscles become jagged or dentated is, that chloride of sodium often induces a similar change of form in normal blood, and that a great concentration of the intercellular fluid promotes the formation of such forms; thus a drop of blood, when it has remained on the stage, and the water has in part evaporated, exhibits

these jagged corpuscles; we usually observe the same appearance in the very saline sputa of catarrhal and phthisical patients, if hæmoptysis be present.

In the *portal blood* of an animal, immediately after it has been killed, we not unfrequently find (according to Schmidt) distorted and jagged bodies of this nature, but they do not occur in the blood of the hepatic veins; this difference may possibly depend on the difference in the quantity of salt contained in the serum of the two kinds of blood; for the serum of the portal blood is richer in chloride of sodium than that of other venous blood, even when the former is of comparatively low density.

With regard to the different substances which simultaneously act on the form of the cells and the color of the blood, we shall here only briefly give the results which we have ourselves obtained, since the statements of different authors present great discrepancies in many points, as might easily be expected.

Amongst the most striking of these phenomena is the expansion of the corpuscles and the simultaneous darkening of the blood on the addition of the various quantities of *water*. The swelling of the lenticular blood-corpuscles is proportional to the quantity of water which is added; they swell, however, in one diameter more than in the other; their concavity on each side disappears, and is replaced by a convexity, so that finally they become converted into spherical vesicles. These often appear to the eye smaller than the pre-existing disks, since it is little more than their transverse diameter that is enlarged, while their long diameter is diminished, if at least only small quantities of water are added. The corpuscles are then very similar to fat-globules, except that they are less glistening and less distinct in outline, as if they had been faintly breathed upon. When the corpuscles have absorbed a large quantity of water, their coefficient of refraction approximates so closely to that of the intercellular fluid, that they can no longer be perceived through the microscope. By the addition of salts to this fluid the blood-corpuscles may again become apparent in their normal form; for the most part, however, they then appear distorted, jagged, or star-shaped. If the blood has been treated with a very large quantity of water, the cell-wall completely bursts, and of course no addition of salts can then restore the integrity of the corpuscles; they then form transparent, granular conglomerations, which may be rendered visible by being treated with a watery solution of iodine, as they then assume a brown color. Blood-corpuscles which have escaped destruction may also be made again visible in watered blood, since the solution of iodine contracts the cell-wall, and gives it a yellow color. The more we add water to whipped blood, the *darker* it becomes in reflected light, but at the same time it becomes *translucent*; while the addition of salt renders the fluid turbid and again opaque, and communicates a light-red tint to it,—a fact whose physical explanation does not require notice in the present place.

The following experiments refer solely to calves' blood. The saline solutions were for the most part applied in the state of saturation at  $+15^{\circ}$ .

In relation to the action of *ether*, Nasse remarks that it renders the

blood-cells smaller and paler, and he believes that a great part of the pigment is extracted by it. The mere results of my experiments on this subject are as follows:

When 100 volumes of blood were shaken with 4·8 volumes of ether, no visible darkening of the blood could be detected; the ether did not again separate from the blood; the blood-corpuscles preserved their form. After 18 hours they slightly sank, but the serum was not yellower than that of calves' blood in general; many corpuscles were then spherical, and some were distorted and had partially lost their sharpness of outline.

On shaking 100 volumes of blood with 8·1 volumes of ether, the blood became decidedly darker; the ether, however, in this case, did not again separate; most of the colored cells disappeared, but those which could still be detected were sharply outlined, spherical, and faintly clouded on their surface; the colorless cells were very distinct.

On mixing 100 volumes of blood with from 12·4 to 24·6 volumes of ether, a dark brown-red, transparent fluid was obtained; here also no ether appeared on the surface, but there was a separation of a light yellowish sediment, which, when examined under the microscope, presented the appearance of coagulated matter (shreds of the membrane forming the cell-walls); only isolated colored corpuscles were seen, and they were pale and distended so as to resemble fat-globules; the colorless cells were as distinct as if the blood had been treated with water.

When equal volumes of blood and ether were mixed, the fluid became very dark, but highly transparent; and standing, a great part of the ether again separated from the blood; here, also, there was a deposition of yellowish flocculi; under the microscope the colorless cells appeared very distinct, but there were scarcely any trace of perfect colored corpuscles; moreover, many large *white* globules of ether were seen in the yellow fluid; the ether collected, after 18 hours, from the watery fluid was colorless even after repeated shakings; from this I drew the conclusion that the ether had not extracted much fat containing hæmatin from the blood-cells.

Salts, such as the sulphates of soda and potash, nitrate and chlorate of potash, and similar compounds, resemble one another considerably in their action; we shall consequently limit ourselves to noticing the relations of the following, which, in this point of view, may be regarded as representatives of the neutral salts of the fixed alkalies.

On mixing 1 volume of blood with 0·8 of a volume of a solution of *nitrate of soda* (saturated at 15°), a light vermilion-colored opaque fluid resulted, in which the blood-corpuscles were strongly contracted in their centre, and had a biscuit-like<sup>1</sup> or drumstick-shaped form. After 24 hours (at 12°), the corpuscles had sunk to the extent of 1·22d of the volume of the fluid; the serum did not separate very completely from the clot, and the whole of it had a somewhat reddish tint; the color of the whole blood had again become somewhat darker, so as to resemble that of unmixed blood; the blood-corpuscles presented very great differences in size and form, and were spherical, angular, elongated, and jagged.

When 100 volumes of blood were mixed with 64·7 volumes of a solu-

<sup>1</sup> [*Backschüssel-biscuit* in the German.—G. E. D.]

tion of common *phosphate of soda*, the resulting fluid was of a light vermillion color, and after 45 minutes the corpuscles began to sink; these were strongly contracted and biscuit-formed; after 23 hours the colored cells had sunk to the extent of about 1-16th of the volume of the fluid; the serum was perfectly colorless, and the clot was of a bright scarlet tint; the corpuscles were still strongly contracted.

On mixing 1 volume of blood with half a volume of a solution of *protocarbonate of soda*, a very light vermillion-colored fluid was obtained; in the course of 40 minutes the corpuscles had distinctly sunk; they were considerably contracted. After 24 hours they had sunk through 1-15th of the volume of the fluid; the color of the blood was very dark; the serum was reddish, imperceptibly verging into the clot, and very tenacious and viscid; the blood-corpuscles were spherical, pale, and clouded.

On mixing 1 volume of blood with 0.7 of a volume of a solution of *bicarbonate of soda*, a very light vermillion-colored fluid was obtained; the blood-corpuscles were very much contracted, and, after 35 minutes began to sink. After 24 hours the color was still of an equally light red, the blood-corpuscles had sunk to the extent of about 1-10th of the volume, and the serum was clear and colorless.

1 volume of blood mixed with 0.8 of a volume of a solution of *ferrocyanide of potassium*, presented precisely the same characters as the preceding mixture; the corpuscles began to sink after 50 minutes. After 18 hours about 1-18th of the volume of the serum was clear and colorless.

1 volume of blood, on the addition of 0.7 of a solution of *borax*, became of a very light red color; the blood-corpuscles were contracted to almost the same extent as by the previous salts, and, after 24 hours had sunk to the extent of about 1-15th of the volume of the fluid; the serum was clear, but reddish.

Blood treated with half its volume of a solution of *iodide of potassium* became of a light vermillion color, and its corpuscles were much contracted and biscuit-shaped; they began to sink in the course of an hour. After 18 hours they had sunk to the extent of about 1-25th of the volume of the fluid; the serum was reddish and turbid, and very distinctly separated from the clot; the whole fluid was of rather a darker red than fresh unmixed blood; it was, moreover, gelatinous and ropy; the blood-corpuscles had lost their discoid shape, and were spherical, but were much smaller than previously, and some of them were very much distorted and jagged.

100 volumes of blood assumed a light vermillion color on being mixed with 44 volumes of a solution of *sulphocyanide of potassium*; the blood-corpuscles were contracted, and began to sink in the course of 34 minutes. In 24 hours the fluid assumed a blackish-brown color; the corpuscles had now only sunk through 1-10th of the volume, but at the same time the serum was reddish and transparent; the clot formed a dark blackish-brown, transparent, clear, perfectly liquid mass, in which no morphological element could be recognized with the microscope.

On the addition of 0.6 of a volume of a solution of *chloride of calcium* (1 part of the salt to 12 of water) to 1 volume of blood, a light red color was produced, although not so light as with most of the alkaline

salts; after an hour the blood-corpuscles began to sink and to contract. After 18 hours there was no further trace of sinking; the corpuscles were then enlarged in their long diameter, and very much diminished in thickness, so that they resembled laminae rather than disks; moreover, they were very much distorted, and some of them had a jagged appearance.

1 volume of blood, mixed with half a volume of the solution of *sulphate of magnesia*, became of a light vermilion color, and remained so even after 18 hours; the fluid had then become very ropy; there was very little sinking of the corpuscles, which had assumed a biscuit-like form and had their long diameter increased; their discoid form was somewhat distorted, and they were often a little jagged at the edges.

On treating 1 volume of blood with two-thirds of a volume of a solution of *hydrochlorate of ammonia*, it first assumed a vermilion color, but, after 24 hours, appeared far darker than blood treated with sulphate of soda, although scarcely darker than unmixed blood; after 1 hour and 5 minutes the corpuscles began to sink, but after 10 hours, there was no true separation of serum; on the surface the mixture was red, and only slightly transparent; it was, moreover, very ropy. The corpuscles were spherical, and smaller in diameter than the original disks.

1 volume of blood, when mixed with half a volume of solution of *cane-sugar* (1 part of sugar to 22 parts of water), became of a somewhat lighter red color; the blood-corpuscles were moderately contracted, and began to sink in an hour and a quarter, the sinking extending to 1-16th of the volume in 18 hours; the serum was perfectly clear and colorless; the clot was of a somewhat lighter color than that of ordinary blood, and the corpuscles were still moderately contracted.

1 volume of blood, on the addition of 0.7 of a volume of solution of *gum-arabic* (1 part in 20 of water), became very dark, and the blood-corpuscles were distended and almost spherical; they began to sink in three-quarters of an hour, and after 18 hours, had sunk to the extent of 1-40th of the volume; the blood had a blackish-red color, and was very tenacious.

100 volumes of blood mixed with an aqueous solution<sup>1</sup> of *arsenious acid*, assumed a somewhat light red color; the blood-corpuscles were unchanged, and, after 24 hours, had sunk to the extent of 1-10th of the volume of the fluid; the serum was then red, and the blood-corpuscles were spherical and had no central shadow; several, that were lying on their edge, were reniform; all were increased in thickness.

1 volume of blood, when mixed with half its volume of extremely diluted *hydrochloric acid* (1 part of hydrochloric acid to 532 of water), became very dark; the blood-corpuscles were not much affected; they were all a little thicker than usual, and those lying on their edge were baton-shaped.

On mixing 1 volume of blood with 0.001 of a volume of *caustic ammonia*, there was scarcely any change of color, and the blood-corpuscles were not visibly altered; after 24 hours they sank to the extent of

<sup>1</sup> [The number of volumes of the solution of arsenious acid, and its strength, are omitted by the author, apparently by an oversight.—G. E. D.]

1-100th of the volume; the serum was then red, and the corpuscles a little distended.

The *caustic alkalies*, and several *organic acids*, as for instance, acetic acid, convert the blood into a blackish-brown, thick, tolerably consistent jelly; and at the same time distend the corpuscles, and distort or destroy them.

We learn, from the observations of Harless, that the primary action of oxygen and carbonic acid on the colored cells, is also of a mechanical nature; but this author has shown, by his variously modified experiments, that these gases likewise exert a chemical influence on every molecule of the blood; thus he found, for instance, that when we allow oxygen and carbonic acid to act alternately on the red cells, they become gradually destroyed, the destruction being usually completed after the ninth or tenth exposure to the action of the gases—an experiment which is obviously of the highest importance in connection with the colored cells in the circulating blood. We should, therefore, at all events, be going too far if, on the above-mentioned grounds, we should ascribe the influence of oxygen or of the gases generally on the color of the blood, solely on the changes in the form of the blood-corpuscle which they induce. The primary action of the oxygen may always be a physical one, like that of the salts; but these also act mechanically only at first; they almost all, as we have already seen, communicate a light-red color to the blood in the first moments of their action; after a longer or shorter period (varying in the case of different salts), they give a more or less dark-red tint to the blood.

It is in the greater or less rapidity of the mechanical action of the salts, that the reason must be sought why a merely chemical action has been ascribed to many of them, when they were only regarded as capable of darkening the color of the blood; as, for instance, to the alkaline carbonates (Mulder and Nasse), the salts of ammonia (Dumas), and the potash-salts, especially nitre (Hünefeld).

Nasse has shown from several carefully conducted series of experiments, that we are by no means justified in drawing any conclusions regarding the action of these substances in the circulating blood of the living body, from their action on fresh blood out of the organism. For a long time he gave to dogs and goats, food containing soda or nitre, but he either observed no action on the coagulation of the blood, or one precisely the reverse of that which he expected. I have made experiments of a similar nature with injections of solutions of *nitrate* and *bicarbonate of potash*; a solution of 30 grammes of nitrate of potash in 200 grammes of water at about 38°, was very slowly injected into the jugular vein of a somewhat overworked horse, which lost little blood by the operation. The operation of venesection was performed a quarter of an hour after the completion of the injection. The blood was rather darker than that which was discharged before the injection, and coagulated more rapidly, but formed a less dense clot and a smaller crust. In a similar manner I injected 30 grammes of bicarbonate of potash dissolved in 180 grammes of tepid water, into the jugular vein of an old but still somewhat powerful horse; seventeen minutes after the completion of the injection, blood was taken from the jugular vein of the oppo-



site side; this blood was much darker than that which escaped before the injection; the blood-corpuscles sank much more slowly, the crust was less thick, and the clot easily broken down. In the latter case the change which the blood underwent from the decomposition of the bicarbonate of potash may be easily explained; in the circulating blood, all the conditions are present which give rise to a decomposition of this salt into carbonic acid and simple carbonate of potash, namely, a high temperature, and the action of free gases; and the fluid has hence assumed the character of a blood rich in carbonic acid: the dark color corresponds with the accumulation of carbonic acid in the blood; the neutral alkaline carbonates, rapidly as it is separated by the kidneys, had, however, here delayed the sinking of the corpuscles. The action of the free carbonic acid was also shown in the excited and, as it were, intoxicated state in which the animal remained even an hour after the injection. This condition was precisely similar to that which I have repeatedly observed in horses, after allowing them to breathe a mixture of 10% of carbonic acid and 90% of atmospheric air for from 3 to 8 minutes; the pulse increased from 36 to 40 strokes in the minute to 50 and even 54; the eyes of the animal were glistening but steady, its gait was firm, there was rumbling in the intestines, and there were eructations and a great flow of saliva.

That the *bicarbonate of potash* is converted in the blood of living animals into carbonic acid and simple carbonate or sesqui-carbonate of potash, is also obvious from experiments which I have made with frogs. These animals were placed in differently saturated solutions of bicarbonate of potash or soda, and were fixed in such a manner that they could breathe freely, and that the web-membrane of one foot could, at the same time, be observed under the microscope. Within three minutes after the beginning of the experiment, the blood-corpuscles began to accumulate in the smaller capillaries of the web-membrane, while in the larger ones there was as yet no perceptible diminution of the rapidity of the circulation; in from 10 to 15 minutes, however, temporary accumulations and short stoppages were perceptible; at a still later period an oscillation began in these larger vessels, so that it was no longer possible to distinguish in which direction the current was running. As far as was possible, the blood-corpuscles of this frog were compared with those of another (not exposed to the action of a salt), whose web-membrane was simultaneously brought under another microscope of nearly the same magnifying power. Nuclei, which, as is well known, are not generally perceived in the blood-cells of frogs' blood in the act of circulation, here also could not be recognized; but although accurate measurements of the blood-cells within the web-membrane could not be made, yet a comparison of the blood-cells in the two kinds of circulating blood, showed (after a condition of stasis had commenced in the finer capillaries of the web-membrane of the frog placed in the salt), that the corpuscles of the blood in which the alkaline bicarbonate was diffused, were swollen, shortened in their long diameter, and dilated transversely. These phenomena and alterations in the dimensions of the blood-corpuscles were even more distinct in frogs which were gradually suffocated in an *atmosphere rich in carbonic acid*.

In both cases, the blood of the larger vessels and of the heart was not of a brownish-red, but of a purplish color, merging from a cherry-red into an almost perfect violet; the blood-corpuscles without a decided nucleus, exhibited a central and peripheral turbidity, independent of the arrangement of the microscope; some were enlarged in diameter and volume. On the addition of bicarbonate of potash to the blood of the frog treated with carbonic acid or the alkaline bicarbonate, the fluid exchanged its purple color for a light vermilion tint; the blood-cells were, however, so contracted that when seen under the microscope they resembled crumpled elliptic laminae, or wrinkled and stippled shreds; their transverse diameter was so diminished that it was scarcely measurable; the nuclei were distinctly visible; they did not, however, present the ordinary form, but occurred as dark granular heaps, which distinctly resembled bone-corpuscles. In both cases the serum separated very completely from the clot; both kinds of blood restored the blue to reddened litmus paper, but only that of the frog which had been exposed to the action of the salt reacted on turmeric paper. The heart of the killed or asphyxiated animals exhibited the singular phenomenon, that, when pinched with forceps, it was thrown into a state of rigid spasm, and by discharging its blood, became perfectly white. In the frogs which had breathed carbonic acid, the lungs were extraordinarily distended, bloodless, and almost colorless; while in the frogs submitted to the action of the salts, they were collapsed and of a cherry-purplish color. In a saturated solution of the alkaline bicarbonates, the frogs died in five minutes; while in a moderately diluted solution, they often remained alive for an hour and a half.

When frogs were treated in a precisely similar manner with solutions of *alkaline protocarbonates*, stoppages of the blood-current in the capillaries were also very soon observed, but no change could be perceived in the dimensions of the blood-cells (either augmentation or diminution of volume); by any possible comparative measurements; the capillaries were, however, very much filled with blood-corpuscles; the intercellular fluid appeared to be diminished, and stasis to be thus induced, precisely as occurs in the phenomena of inflammation; here also there were no nuclei to be perceived. The blood of the larger vessels had not the slightest tint of violet, but was of a pure brownish-red color; its corpuscles were, however, collapsed, in folds, strongly granular, and presented a dull granulated nucleus; on the addition of an alkaline protocarbonate, they became still more contracted, and the nuclei stood out distinctly as minute accumulations of sharply projecting granules, the entire cell having a shred-like and folded appearance, and being dotted with tolerable regularity on its border; on exposure to the air, the dark reddish-brown clot assumed a light-red color. The lungs were moderately collapsed, and of a brownish-red color; the heart, on being touched, was not thrown into a state of rigid spasm, but was excited to active contractions.

In frogs narcotized with *ether*, and observed in a similar manner, some striking phenomena, very different from those hitherto mentioned, were noticed; here, during the gradual action of the ether, stoppages in the circulation of the web-membrane were remarked, but instead of an

accumulation of blood in the smaller capillaries, many of them were perfectly devoid of colored cells, so that in some nothing but a few scattered colorless corpuscles could be recognized; no blood-corpuscles any longer passed from the larger vessels into the apparently empty spaces; the diameter of the smaller capillaries was obviously so diminished that no red blood-cells could any longer enter them, and those which had been contained within them, streamed forth from their mouths, which were distinctly visible; no change could be observed in the blood-corpuscles themselves. The blood of the larger vessels was of a dark-red color, merging into violet; its corpuscles during the first few moments were normal and without a nucleus, but on exposure to the air they soon became distorted and indistinct. The lungs were usually (but not always) filled with air, and very much expanded. It is perhaps deserving of mention that after etherization the muscles were always found in a highly relaxed condition, while after the application of carbonic acid or alkaline carbonates, there was constantly tonic spasm, and the muscles, after death, were found (as has been already mentioned) in a state of rigid contraction; if we regard this phenomenon as a consequence of irritation or paralysis of the spinal nerves, we should have observed paralysis of the vasomotor nerves of the web-membrane in rigor of the muscles, and irritation of the vasomotor nerves in paralysis of the spinal nerves.

We should scarcely have described so fully, in this place, the relations of the above-named substances in the blood of living animals, if we had not wished at the same time to use these experimental researches as a caution against the too hasty conclusions that have been drawn from the action of various chemical substances on the blood-corpuscles and other elements of the blood, with the view of elucidating pathological and pharmacological processes. If in the more recent and so-called rational pharmacology, we had guarded against such crude chemical explanations, we should have avoided many errors, and kept clear of many absurd physiological fictions.

It is clear, from the preceding remarks, that many of those substances which modify the form of the blood-corpuscles, at the same time exert a chemical action on their walls; but whether they—and more especially the gases—extend their action to the contents of the blood-cells, and especially to the pigments, is a question as yet by no means satisfactorily answered. To judge from the properties of hæmatin, as described in this volume, we should scarcely expect such an action; for we have there seen how indifferent and inaccessible hæmatin is to most chemical reagents; but, on the other hand, it is also manifest that this pigment can hardly exist in the blood-cells in the same state in which it is exhibited isolated by chemists. There is still a perfect absence of definite chemical facts to prove the almost indubitable action of oxygen on the contents of the blood-corpuscles; there are merely a few experiments made by Bruch<sup>1</sup> in support of this view, which has become more than probable from physiological grounds. The pigment itself appears to undergo changes of color by oxygen and carbonic acid; for if strongly watered blood, in whose plasma it may be presumed that the contents

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 1, S. 440–450, and Bd. 3, S. 308–318.

of the blood-corpuscles are diffused, be shaken with carbonic acid gas, its dark color becomes still darker in refracted (or. transmitted) light; that is to say, blood which is merely watered appears, when viewed by transmitted light, of a less deep dark-red color than blood which has been similarly watered and has been impregnated with carbonic acid; we observe the opposite result on treating blood, which has been watered in this manner, with oxygen gas. This may serve to explain why the blood of the portal vein, which is richer in water than that of the other veins, is also of a darker color.

Taking into consideration all these circumstances regarding the mechanical relations of the blood-corpuscles, it follows that a definite tint may be given to the whole blood by the action of very different influences upon the blood-cells, and that in special cases it is often very difficult to decide on which of these often opposite causes the color of the blood in any particular case may depend.

Moreover, there are other physical relations, not directly acting on the blood-corpuscles, which may modify the color of the whole blood. Thus we find the blood of a lighter tint when, in addition to the red cells, it contains a very large number of colorless corpuscles, or of other particles which strongly reflect light; thus Scherer showed that the addition of milk or powdered gypsum made the blood of a lighter red tint; and this is also the reason that we sometimes find the blood, in cases of pyæmia, which abounds in colorless blood-cells, as well as the blood of confirmed drunkards, in which there are innumerable fat-globules, of a comparatively light tint.

We need hardly mention that external influences, such, for instance, as putrefaction, must act on the form of the corpuscles and give rise to chemical changes. Hence we need not wonder at meeting with blood-corpuscles of the most varied forms in the blood of dead bodies or in old exudations. It is, however, only rarely that we can draw any conclusions regarding the pre-existing disease from these forms; for they are not the direct result of a morbid process, but merely the consequence of the chemical or physical changes to which the intercellular fluid is exposed. We must not, therefore, expect any great advantage for medical diagnosis from the microscopic examination of such blood; on the one hand, because such changed forms of the corpuscles never occur in fresh blood (although they were formerly supposed to have been found in the blood in cases of typhus); and on the other, because blood obtained from the dead body always rapidly undergoes essential changes from external influences.

Having thus considered the physical characters of the blood-corpuscles, we now proceed to the investigation of their *chemical constituents*. This is a subject on which there is still much that is obscure. The microscopic examination of the blood has certainly taught us that its pigment is limited to only the colored cells; Berzelius has further shown that in these cells there is contained an albuminous fluid, differing, however, from albumen; which he named globulin, and expressed his belief that the phosphorized fat was in all probability only contained in the blood-cells. The same chemist likewise indicated the way by which the blood-

corpuscles might be separated from the intercellular fluid, or by which, at all events, they might be obtained free from the constituents of the serum, although with the loss of several of their own essential constituents. Dumas and Figuier were the first to apply this method to actual practice; and the former, by this means, was enabled to submit to an elementary analysis the dried fragments of blood-corpuscles; but, from the very nature of the case, all these investigations could lead to very few conclusions regarding the true and essential constituents of these colored cells; for we were investigating either the blood-cells mixed with intercellular fluid, or merely the cells more or less completely freed from all soluble substances (penetrating the cell-walls). We are indebted to the ingenious and careful investigations of C. Schmidt for a more definite knowledge regarding the composition of the contents of the blood-corpuscles, and the nature of the individual substances occurring in them. We shall see that in this discovery of Schmidt's lies the nucleus of all our knowledge and theories regarding the chemico-physiological importance of the blood-cells.

Berzelius<sup>1</sup> had already found that the corpuscles were the cause of the blood's redness, and were not dissolved by salts having an alkaline basis, or by sugar, and that we had in this way a means of separating in some measure the blood-corpuscles from the intercellular fluid. Figuier<sup>2</sup> was the first who employed this means to obtain a quantitative determination of the blood-corpuscles, regarding which we shall speak more fully presently. Since, however, in attempting to separate the blood-corpuscles in this way (a solution of sulphate of soda is what is commonly used for the purpose) from the constituents of the intercellular fluid, it very soon becomes apparent that these particles, on the one hand, become agglutinated to, and stop up the filter, and on the other, that they become so changed that they pass through it, Dumas<sup>3</sup> recommended that oxygen should be continuously passed into the fluid lying in the filter, while, at the same time, a solution of Glauber's salts should be constantly allowed to drip into it. The blood-corpuscles obtained in this manner, and containing Glauber's salts, are dried, extracted with ether and boiling alcohol, and finally freed by boiling water from the sulphate of soda and other soluble constituents. By submitting to ultimate analysis this residue of the blood-cells freed from serum, Dumas found that both in men, dogs, and rabbits, after deducting for the ash, there was the constant ratio of from 55.1 to 55.4% of carbon, 7.1% of hydrogen, from 17.2 to 17.5% of nitrogen, and consequently from 20.2 to 20.6% of oxygen.

C. Schmidt<sup>4</sup> exhibited in a similar manner the coagulable and insoluble parts of the blood-cells, and found their specific gravity before the abstraction of their iron to be 2.2507; but after the abstraction of the ash and iron only 1.2090. The same author found that 100 parts of this dry cell-residue contained on an average 87.59 parts of globulin and 12.41 of hæmatin. The residue, containing ash, yielded 1.179% of peroxide of iron and 0.126 of earthy phosphates.

In reference to the *cell-wall* of the red corpuscles, most French

<sup>1</sup> Lehrb. d. Ch. Bd. 9, S. 74. (4te Aufl.)

<sup>2</sup> Ann. de Chim. et de Phys. 3me Sér. T. 11, p. 508.

<sup>4</sup> Ann. d. Ch. u. Pharm. Bd. 61, S. 156-167.

<sup>3</sup> Ibid. T. 17, p. 542.

chemists, even to the most recent time, have held that this membrane was fibrin, in accordance with the old view regarding the coagulation of the blood. Denis and Lecanu have attempted to demonstrate the presence of fibrin in the blood-corpuscles by triturating them with salts, viz., nitrate of potash and chloride of sodium; Virchow, who has repeated these experiments, has however shown that the small membranes observed by these authors are nothing more than the folded and adhering walls of the blood-corpuscles, which, under the microscope, in consequence of the pressure and the crushing of the glass covering the object, often acquire the appearance of Nasse's fibrinous flakes; Virchow, however, very correctly remarked that the solubility of these membranes in a solution of nitre, and their swelling in acetic acid, by no means prove their identity with fibrin: moreover I was unable to obtain a trace of coagulable matter, or of matter precipitable by acetic acid, from the cell-membrane of the corpuscles of the blood of horses and oxen by prolonged digestion with a solution of nitre. Mulder regards the cell-wall as binoxide of protein; but the properties of the remains of the cell-walls obtained by treating blood with water by no means coincide with those of Mulder's binoxide of protein; they are far more difficult of solution in acetic acid and in the alkalis than the latter; and in these membranes I have not been able to detect any trace of sulphur, which, as is well known, is contained in binoxide of protein. Moreover, Mulder has not demonstrated the presence of this substance by direct experiments, but was merely led to this view by the following consideration: on their passage through the pulmonary capillaries, the blood-corpuscles become invested with a thicker layer of this binoxide, in consequence of which the blood-pigment appears of a lighter red color, as if seen through ground glass, and hence the lighter red tint of arterial blood; the central depression of the colored cells also bears out this view, since the inflammatory crust in which, as is well known, there is much binoxide of protein, has also a great tendency to exhibit a similar depression or concavity.

It is very probable that the cell-walls of the corpuscles even of the same blood have not a precisely identical composition; at all events we see that the colored cells of the same blood are, as a general rule, very unequally acted upon by the same reagents; if, for instance, we allow water, dilute acids, ether, or dilute alkaline solutions, to act on the blood-corpuscles, we perceive that the work of destruction does not by any means proceed uniformly; thus some do not disappear even when the blood is very much diluted with water; these we consider to be the younger cells, while those which are easily destroyed are regarded as the older blood-corpuscles; for it is believed that the capsule of the colorless corpuscles, from which the colored cells at all events in part proceed, retains for some time its former chemical nature, even when pigment has become formed within the cell. The cell-wall, which so rapidly disappears from our sight under the microscope, is, however, actually dissolved by very few of these reagents; it only passes into a gelatinous or rather a mucus-like condition, in which its coefficient of refraction is nearly the same as that of the plasma; we arrive at this conclusion not merely from the experiment to which reference has been

frequently made, by which the cell-wall may again be rendered visible either in all its integrity or at all events in fragments by solutions of salt, iodine, &c., but also from the viscosity and tenacity which are imparted to the blood by the addition of certain substances, as dilute organic acids, alkaline carbonates, iodide of potassium, hydrochlorate of ammonia, &c. If blood which has been thus modified be saturated with acids or alkalies, or if a solution of iodine or of sulphate of soda be added to it, the walls of the corpuscles again become apparent, and the blood at the same time loses its acquired viscosity. Moreover neither the intercellular fluid nor the serum is reduced by the above means to such a viscid or tenacious condition, which must therefore be dependent on the blood-corpuscles: further, mucus which had become swollen in water becomes condensed by the same means, so as to be less transparent to the unaided eye, appearing almost as if it were coagulated, and exhibiting thread-like streaks under the microscope.

The *globulin*, or coagulable matter contained in the blood-cells, as well as the *hæmatin*, has been fully considered; we shall therefore direct our attention to the other organic substances which must be regarded as essential constituents of the colored cells.

With regard to the *nuclei of the blood-corpuscles*, in a morphological point of view they are of very doubtful importance, since several of our first physiologists (R. Wagner amongst the number) regard the very distinct and often clearly defined nuclei of the blood-corpuscles of the amphibia as products of chemical secretion from the homogeneous cell-contents after death, while others conceive that in the discoid colored bodies in the blood of mammalia and birds they see the nuclei or their remains. But whatever decision may be arrived at regarding the morphological existence of these elements, nothing can as yet be definitely concluded regarding their chemical nature, in the first place because we are altogether unable to isolate them for chemical examination, and secondly, because even if we recognize them as composed of a protein-compound, our knowledge of these substances is still such a vexed question, that it would be impossible to decide whether this nucleus-substance did or did not consist of one of the known and named protein-bodies.

J. Müller, and subsequently F. Simon, regarded the nucleus as fibrin in consequence of its solubility in acetic acid and in alkalies, but unfortunately these properties are not characteristic of the substance to which the term fibrin is generally applied; moreover, my observations coincide with those of Jul. Vogel, who found the nucleus very difficult of solution in acetic acid, and hence I cannot regard it as identical with fibrin. Maitland<sup>1</sup> regards the nucleus as consisting of a peculiar horn-like compound, which he named *nucleine*; Nasse very correctly remarks that the substance which Maitland obtained by washing the clot after the removal of the fibrin at the same time contains the cell-walls of the blood-corpuscles, which at all events preponderate very much over the nucleus-substance in question. Hünefeld regards the nucleus as consisting essentially

<sup>1</sup> An Experimental Essay on the Physiology of the Blood. Edinburgh, 1838, p. 27.

of fat; that fat is abundant in the blood-cells will be immediately shown; but it is scarcely necessary to mention that in the process of exhibiting these nuclei, the fat must always become mixed with them, and consequently must always form the larger part of the object of investigation.

It has been already mentioned that a considerable part of the *fat* of the blood is accumulated in the blood-cells. Berzelius thought it probable that the so-called phosphorized fat might be chiefly contained in the blood-corpuscles. I have at all events found this view to be so far correct that the fat extracted by ether from the blood-corpuscles of the ox (obtained by means of sulphate of soda, according to Dumas's method) yielded about 22% of ash, which had an acid reaction, and consisted essentially of acid phosphate of lime. Since, however, at the present day we are justified in questioning the existence of such a phosphorized fat as was formerly supposed to exist, the idea suggests itself, that what we here meet with is the glycerophosphoric acid discovered by Gobley in the yolk of egg. In the dry blood-corpuscles of the ox I found on an average 2.249% of matter extractable by ether. We must, however, not omit to mention that the blood-cells of arterial are poorer in fat than those of venous blood; thus in the corpuscles of the arterial blood of a horse I found only about half as much fat as in those of its venous blood; in the latter the amount being 3.595%, and in the former 1.824% of the dry prepared corpuscles.

The so-called *extractive matters* of the blood cannot be accurately indicated, since they are substances of which we have no knowledge; but this much is established from the few investigations which I have made on this subject, namely, that most of such substances pertain to the serum and not to the blood-cells. While 100 parts of the solid residue of the serum contain about 8 parts of extractive matters free from saline constituents, 100 parts of the solid residue of the cells of the same blood (calculated from the analysis of the clot) do not contain 6 parts of such substances.

In regard to the *mineral constituents* of the blood-corpuscles, very different views have been held regarding them, which are all almost equally removed from the truth; this observation, however, does not extend to the iron. It has either been believed that all the salts which we find, or presume to exist, in the serum, must also be contained in the blood-cells, or it has been assumed that at all events the soluble salts, especially the chlorides of sodium and potassium, are altogether excluded from the cells. Although neither of these views is yet generally adopted, and in the absence of all means of deciding between them, we refrain from a definite opinion, yet at all events the ideas at which we arrive from analyzing the blood point only to these two modes of considering the subject. We are indebted to the unremitting investigations of C. Schmidt for a series of facts which prove that, in reality, soluble salts are also contained in the moist blood-cells, that these salts are by no means perfectly identical with those which we find in the serum, and finally, that their quantity is far smaller than it must be if the water of the blood-corpuscles contained exactly the same amount of saline matter as the water of the serum.



We need only institute a comparison between good analyses of the serum and of the clot of the same blood, and by a most simple calculation subtract the soluble salts occurring in the serum (surrounding the cells) from the sum of the soluble salts of the clot, to convince ourselves that by far less of such salts can be contained in the blood-cells than in the serum, but at the same time that the salts cannot pertain to the enclosed serum alone.

Thus, for instance, in the serum of the venous blood of a horse I found 0.835% of salts (soluble and insoluble), and in the moist clot of the same blood 0.819% of salts (including peroxide of iron); deducting the 0.114 of peroxide of iron found in it, there remains 0.705% of mineral substances; if now we suppose by way of illustration, that the clot is so loose that it contains enclosed within it one-third of its weight of serum, then we should have to deduct 0.273 for the enclosed serum from the 0.705 of salts, and there would then remain only 0.432 of salts for the 66.667 of blood-cells (which correspond with two-thirds of the original weight of the clot); hence, in 100 parts of moist blood-cells, there would be only 0.648 of salts. If, however, we assume that the blood-corpuscles lie so closely together, that the serum which they enclose amounts to only one-fifth of the weight of the clot, then, since 16.667 parts of serum contain 0.137 of salts, there will remain only 0.568 of salts for the 83.333 parts of blood-corpuscles; that is to say, in 100 parts of blood-cells, there will be 0.681 of salts. As we shall presently show, Schmidt has now found out a method of discovering with tolerable accuracy the quantity of the serum enclosed in the clot, and hence, of calculating the mineral constituents occurring in the moist blood-cells.

Although we are not able to calculate the quantity of the mineral constituents contained in the fresh blood-cells, the questions still remain to be answered whether there are certain salts which especially accumulate in the cells, and if so, which they are. These questions have also been answered by C. Schmidt; for he has discovered that the fluid of the blood-cells (that is to say, the water contained in the blood-corpuscles) contains in addition to the organic matters, a preponderance of phosphates and potash-salts; so that, consequently, the phosphate of potash and the greater part of the chloride of potassium pertain to the blood-cells, whilst the chloride of sodium, with a little chloride of potassium and phosphate of soda, is found in the plasma (serum + fibrin). In the plasma, the organic materials are combined only with soda, while in the blood-cells, the fatty acids and the globulin are combined both with potash and soda.

C. Schmidt, in analyzing a specimen of blood, which contained 396.24 p. m. of blood-cells and 603.76 p. m. of intercellular fluid, found 1.353 of chloride of potassium and 0.835 of phosphate of potash in the former, while there were 3.417 parts of chloride of sodium, besides 0.267 of phosphate of soda and 0.270 of chloride of potassium in the latter.

Schmidt has examined and tabulated the relations between potassium and sodium, and between phosphoric acid and chlorine in the blood-cells and in the intercellular fluid in several of the mammalia.

The following table contains the chief results of his observations:

100 PARTS OF INORGANIC MATTERS :								
GENUS.	BLOOD-CELLS.		PLASMA.		BLOOD-CELLS.		PLASMA.	
	K	Na	K	Na	PO	Cl	PO	Cl
Man, (mean of 8 exps), .	40.89	9.71	5.19	37.74	17.64	21.00	6.08	40.68
Dog, . . . . .	6.07	36.17	3.25	39.68	22.12	24.88	6.65	37.31
Cat, . . . . .	7.85	35.02	5.17	37.64	13.62	27.59	7.27	41.70
Sheep, . . . . .	14.57	38.07	6.56	38.56	8.95	27.21	3.56	40.89
Goat, . . . . .	37.41	14.98	3.55	37.89	9.41	31.73	5.90	40.41

These results coincide with those of Nasse, who found the most phosphates in the blood of those animals which were distinguished for the abundance of their blood-corpuscles, namely, swine, geese, and hens; in sheep and goats, on the other hand, in whose blood he found comparatively few corpuscles, he also found the least phosphates. On another occasion, Nasse<sup>1</sup> has also expressed the opinion that the phosphates must be principally contained in the blood-corpuscles.

In man, as we see, this difference is the most obvious; in the carnivora it is most marked in the acids; and in the herbivora, in the alkalis. Schmidt adds, that the nature of the food which the animal may take, or variety of race in the case of man, exerts no influence on these relations.

*Earthy phosphates*, as we have already mentioned, also occur in the blood-cells, but both relatively and absolutely in far less quantity than in the intercellular fluid.

In the blood-cells of 1000 parts of blood, Schmidt found only 0.086 of the phosphates of lime and magnesia, while in the intercellular fluid he found 0.332; or in 1000 parts of blood-cells, 0.218, and in 1000 parts of intercellular fluid, 0.550 of earthy phosphates.

The *iron* of the blood pertains, as is well known, almost entirely to the hæmatin of the blood-cells; since the quantity of iron in the ash, when compared with the number of colored blood-cells, is somewhat variable, we conclude, as has been already stated, that the quantity of hæmatin must consequently vary in the blood-cells.

We have seen that the blood-corpuscles obtained from the hepatic veins contain less peroxide of iron than those from the portal vein. Schmidt found an excess of iron in the blood-cells in the hydræmic conditions, and hence he concludes that in these cases the blood-cells have become poorer in globulin, but not richer in hæmatin; he believes, however, that we may calculate the hæmatin from the quantity of iron found in the ash; and this conclusion certainly seems justifiable if we had to do with the pure hæmatin of the chemist, which contains 6.6% of iron; but we must take into consideration that the hæmatin, in all probability, does not start, Minerva-like, into perfect being, but that, almost to a certainty, it is gradually formed, even as it is gradually destroyed; to which it must be added that we are already acquainted with (artificially prepared) non-ferruginous hæmatin; and how, then, can we tell whether, in some

<sup>1</sup> Handwörterbuch der Physiologie, Bd. 1, S. 165.

organ or other, we may not discover hæmatin either altogether free from iron, or, at all events, poor in that constituent?

Schmidt has convinced himself, by several series of experiments, that the clear serum of the blood of oxen, sheep, swine, horses, dogs, cats, rabbits, and hens, is perfectly devoid of iron. (Nasse had previously found this to be the case.)

In 100 parts of dry blood-corpuscles (determined according to the method of Prevost and Dumas) Schmidt found the following proportions of iron: in man, 0.4348%; in the ox, 0.509%; in the pig, 0.448%; and in the hen, 0.329%.

The *gases* of the blood, carbonic acid, nitrogen, and oxygen, are also for the most part contained in the blood-corpuscles. It has been ascertained by Davy, Nasse, Scherer, van Enscht, Magnus, and others, that the serum possesses in a far less degree than the defibrinated blood the capacity of absorbing oxygen and carbonic acid, and I have convinced myself that at least twice as much air is developed from a volume of whipped air *in vacuo* as from an equal volume of serum that has been strongly stirred or shaken with atmospheric air. Van Maack has found that a solution of hæmatin possesses a decided power of attracting oxygen; and Scherer has not only convinced himself of the accuracy of this observation, but at the same time ascertained that a little carbonic acid is developed after the absorption of the oxygen.

Davy and Berzelius believed at one time that they had convinced themselves of the presence of *free gases* in the blood, but subsequently retracted this view; after this period, the results of different experimentalists were very discordant, some being in favor of and others opposed to the presence of gases in solution in the blood. The question was, however, decided about ten years ago, by the experiments of van Enscht, Bischoff, John Davy, and especially Magnus, which showed that free gases are contained in solution in perfectly fresh blood, both arterial and venous; more recently, and by means of simpler experiments, Magnus<sup>1</sup> has confirmed his former observation, that, in addition to carbonic acid, both free oxygen and nitrogen occur in the blood. According to the earlier investigations of Magnus, arterial and venous blood contain nearly equal quantities of nitrogen; in the former, the oxygen is to the carbonic acid (by volumes) as 6 : 16, and in the latter as 4 : 16; hence, therefore, there is relatively more oxygen in arterial than in venous blood. His more recent experiments<sup>2</sup> determine not merely the ratio of the volumes of the gases to one another, but also to the volume of the blood; they show that at all events in the blood of calves, oxen, and horses, there are always in solution from 10 to 12.5% (by volume) of oxygen, and from 1.7 to 3.3% (by volume) of nitrogen. According to an experiment of Magendie's, venous blood contains 78, and arterial only 66% (by volume) of carbonic acid. The oxygen of the blood may also be almost entirely extracted *in vacuo*, as well as expelled by other gases, as for instance, hydrogen and carbonic acid; whence it is sufficiently clear that it is only mechanically absorbed in the blood, and not in a state of chemical combination. Since the blood, according to the experiments of Magnus, is capable of absorbing  $1\frac{1}{2}$  times its volume, or 150% of carbonic acid, it

<sup>1</sup> Pogg. Ann. Bd. 36, S. 685 ff.

<sup>2</sup> Ibid. Bd. 56, S. 177-206.

may, at first sight, appear strange that the circulating blood is not found to be more impregnated with carbonic acid, and that in respiration there is only little more oxygen absorbed than carbonic acid given off; but when we consider that in respiration the relations of the concurrent gases are altogether different from what they are in our experiments (in which we shake pure atmospheric air or pure carbonic acid with the blood), this difficulty is at once removed.

In connection with the occurrence of free oxygen in the blood, there is an important, and indeed even yet a scarcely decided question—whether, at all events, a portion of the oxygen that finds its way through the lungs into the circulation, does not at once chemically combine, in the arterial system, with some of the constituents of the blood. Marchand attempted to decide this question by certain experiments, and Magnus by a calculation based on established facts. Marchand believed that if blood containing no carbonic acid produces no carbonic acid by the direct influence of oxygen, the oxygen can only be mechanically absorbed: now he found, in point of fact, that fresh blood after its carbonic acid had been removed, was as incapable of developing the slightest trace of carbonic acid, when a stream of oxygen was passed through it, as the serum of the blood, egg-albumen, solutions of blood-corpuscles, &c., when similarly treated: but independently of the circumstance to which we have already referred, that van Maack and Scherer have actually observed the exhalation of carbonic acid from hæmatin after its previous absorption of oxygen, nothing more is proved by Marchand's experiment than that oxygen can be absorbed by the blood without giving rise to the formation of carbonic acid; but it is still quite possible that one or other of the constituents of the blood becomes more highly oxidized without any separation of carbonic acid, since a development of this gas does not of necessity follow every oxidation of an organic body. The mode of calculation adopted by Magnus, would be more convincing, were it not that the numbers on which it is based, rest on too uncertain determinations. If, for instance, *about* 13 Paris cubic inches of oxygen make their way into the blood of an adult man in one minute—if, further, *about* 10 pounds of blood pass through the lungs in the same interval—then, considering that *about* 11½ of oxygen are found in the blood of the horse, it follows that *about* half the oxygen which Magnus found in arterial blood has been absorbed from the venous blood, so that, according to this, the former would always lose about half its free oxygen in the capillaries. The preceding statement and the above-mentioned facts afford a sufficient proof that the greater part of the oxygen absorbed in the lungs, exists in a state of freedom in the blood; but it seems to us not at all indubitably established that no portion whatever of the absorbed oxygen enters into chemical combination with one or other of the constituents of the blood, even in the heart and arteries, since such a combination is believed to take place in the capillaries.

Liebig<sup>1</sup> has recently adduced new and striking proofs in the latter view. Water absorbs only 0·925% of its volume of oxygen, whilst, according to Magnus, from 10 to 13% may be taken up by the blood; this greater force of absorption in the blood can only depend upon certain

<sup>1</sup> Chem. Briefe. 3 Aufl. S. 419–423 [or Letters on Chemistry, 1851, pp. 332–335].

constituents, and principally, as we know, upon the red corpuscles; only from 1-14th to 1-11th of the oxygen which is absorbed by the blood, and which varies from 10 to 13%, can be absorbed mechanically, that is to say, by the water, or can consequently exist free in the blood; the remaining oxygen, that is to say from 13-14ths to 10-11ths, must therefore be fixed by certain blood-constituents; but this is only conceivable through the agency of some chemical attraction, however slight that may be. The chemical combination of oxygen with the constituents of the blood may be very loose and entirely analogous to the combination in which the carbonic acid exists in the blood, as already described. The mechanical solution of a gas is entirely dependent upon the pressure which it has to sustain; if a definite quantity be absorbed independently of external pressure, and if this amount stands in a direct proportion to any definite constituent of the fluid, the increase in the absorbing power of the fluid cannot be referred to any cause but chemical attraction. Although, in the case of the oxygen, we are not so well acquainted with the matters which retain it, as with those which are able to fix the carbonic acid in the blood (alkaline carbonates, phosphates, &c.), the proposition is almost equally established for both cases, that the excess of carbonic acid and oxygen which the blood is able to absorb beyond the amount which corresponds to the quantity of water which it contains, must be present in the blood in a state of chemical combination. We have already endeavored to show that the possibility of breaking up such an unstable combination by the aid of other indifferent gases (as hydrogen, &c.) furnishes no evidence against the fact that the expelled gas has been chemically combined. Liebig is therefore certainly in the right when he advances the proposition, that a gas can only be considered as mechanically absorbed when its quantity increases and diminishes in proportion to the external pressure. We think we are justified in concluding with Liebig, that the quantity of oxygen which may be absorbed by the blood is constant in amount, and to a certain extent independent of external pressure—an opinion which is based partly upon the fact, that the respiratory process is carried on nearly the same, both at very great heights and at the level of the sea, and that no more oxygen is absorbed even in an air very rich in oxygen than in the ordinary atmosphere.

In addition to these physical proofs in favor of the chemical absorption of the oxygen in the blood, I may perhaps be permitted to refer to the following experiments, instituted by myself, with the pure crystalline substance of the blood, although they can scarcely be said to furnish any conclusive result. A perfectly limpid saturated solution of pure blood-crystals, which was not precipitable either by nitrate of silver or by basic acetate of lead, and which was of a beautiful pomegranate-red color, was saturated, one part with carbonic acid and another with oxygen; the oxygenous fluid exhibited no remarkable difference of color from the original fluid, which was contained in a similar vessel; moreover, no distinct difference of color could be perceived between the solution which was impregnated with carbonic acid and the normal fluid, or the solution impregnated with oxygen; but the solution of the blood-crystals through which carbonic acid had been passed was somewhat turbid, and exhibited under the microscope large numbers of faintly

granulated flakes. In vacuo the latter fluid developed a very large amount of gas, and retained its turbidity and color; these flakes remained unchanged when seen under the microscope. Both the normal fluid and the fluid impregnated with oxygen remained unchanged both as to color and clearness in vacuo, although they developed relatively less gas. When the solution of blood-crystals was first saturated with oxygen, and then exposed to a stream of carbonic acid, the fluid became turbid without any marked change of color, and exhibited the same flakes under the microscope as the solution which had been treated directly with carbonic acid. If, however, a stream of oxygen be suffered to pass through the fluid, which has been rendered strongly turbid by carbonic acid, it at once becomes perfectly limpid, without exhibiting any perceptibly increased lightness of color. The substance may be again obtained in a crystallized form and unchanged, both from the turbid solution charged with carbonic acid and the clear fluid to which oxygen has been added. This separation of solid molecules by carbonic acid might seem to present a strong indication of a chemical action of the gases, but it could not be made to correspond with the opinion of Bruch,<sup>1</sup> that the normal color of the substance in question is developed in the presence of carbonic acid. Other gases than oxygen and carbonic acid evidently exert a chemical action, like dilute organic acids, alkalies, &c.; this, for instance, is the case with the carbonic oxide, for it not only very considerably darkens the deep red solution of the pure crystals, but it also gives rise to a dark brownish-red coagulum (which exhibits great variety of form when seen under the microscope). Neither solutions of the original color, nor crystals, can be procured from this fluid, either by repeated treatment with oxygen, or with carbonic acid and oxygen. Nitrous oxide renders the red fluid darker, almost brownish-red, and very turbid, so that the microscope exhibits the whole fluid as if it were filled with flakes; neither oxygen nor carbonic acid restores the clearness or the original color of the fluid, for the greater part of the substance crystallizes unchanged. (Nitrous oxide may, therefore, be employed in the place of the oxygen in the mode of preparing the crystalline substance of the blood already described, but it cannot take the place of the carbonic acid). There can scarcely, therefore, be any doubt that the gases (oxygen and carbonic acid) exert a chemical action upon the coloring principle of the blood-corpuscles, as we see from these experiments, as well as from the entire mode of preparation of the crystalline substance; although it would require far more extended investigations to exhibit the true mode of their operation. It would, however, be going too far, were we to conclude that the property which a protein-body exhibits of being modified in its character by oxygen and carbonic acid, is anything peculiar to this substance, or is any special attribute appertaining to this constituent of the blood-corpuscles. For, independently of the fact that, as we have already mentioned, globulin can be completely precipitated from its neutral solutions by carbonic acid, and the precipitate can be again dissolved by a stream of oxygen, certain modifications of the crystalline substance occur, which stand in

<sup>1</sup> Zeitsch. f. rat. Med. Bd. 1 S. 440-450, Bd. 3, S. 308-318, and Zeitsch. f. wissenschaft. Zoologie, Bd. 4, S. 373-376.

precisely the reverse relation to these gases. I have already described that modification of the crystalline substance which can be exhibited by acetic acid and an alkaline salt, and which corresponds with Panum's acid albumen. When the faintly acid solution of this product of metamorphosis is carefully neutralized with a very dilute solution of potash, the substance will be perfectly precipitated; but this precipitate dissolves again in pure water, although not to any great extent, forming a pale-red solution, from which the substance may be so completely thrown down by oxygen or by the action of the air, as only to leave a perfectly colorless fluid over the dirty flesh-colored precipitate. On passing a stream of carbonic acid through this fluid, the precipitate redissolves into a pale-red fluid. The substance may be again precipitated by oxygen, while the solution coagulates on boiling, in the same manner as albumen. These experiments, notwithstanding their isolated character, contribute, together with what we have already stated in the last page, in reference to the influence of the gases upon the formation of the blood-crystals, to strengthen the probability that a chemical action may be impressed upon the main constituent of the blood-corpuscles by the alternating action of oxygen and carbonic acid, although opinions may differ as to whether this is manifested by an oxidation or a reduction, or whether it arise from the simple occurrence of carbonic acid as a conjugated acid, or finally, whether it be referable to a salt-like compound.

Here I cannot refrain from observing that the substance corresponding to the acid albumen has not fallen under my notice as a product of decomposition of the true crystallizable matter of the blood, although Panum is of opinion that the albumen may be decomposed by acids and potash-salts into acid albumen and into another organic substance. No other organic body is formed from the crystalline substance during the preparation of this matter; only a few phosphates separate from it, as is shown by my comparative analyses of the crystalline substance and of its products of metamorphosis, as well as by my investigation of the acid and saline fluid obtained after the removal of the precipitated bodies by filtration. It is only a metameric modification of the original crystalline substance.

I regret that Meckel's paper on hæmatoglobulin, contained in Scherer's Jahresbericht,<sup>1</sup> has only reached me while these sheets were passing through the press. According to Meckel, oxygen changes hæmatoglobulin to a bright-red, and carbonic acid to a bluish-red color. It will be seen from my previous remarks, that I did not succeed with certainty in detecting this or any similar change of color in the pure crystalline substance which I obtained. Moreover, according to Meckel, arterialized hæmatoglobulin is not crystallizable, and only the quantity corresponding to the globulin of venous blood (dem venosirten Globulin) crystallizes from the blood—a fact which it seems to me difficult to prove, as Meckel appears entirely to have overlooked the influence of light upon the formation of crystals. Although it may be *à priori* highly probable that hæmatochlorin and hæmatoïdin appear to be produced from hæmatoglobulin by oxidation, I cannot discover any chemical proof of the fact

<sup>1</sup> Jahresber. d. ges. Med. 1852, S. 95.

of Scherer's report. Meckel also employed a stream of carbonic acid in the crystallization of his hæmatoglobulin, and in the course of his experiments he made numerous valuable observations, which tend to confirm many other researches, more especially those of Kunde and Funke.

In every case the relation of the gases to the blood-corpuscles must be accurately determined by special experiments before a definite view can be formed on the subject.

Before we proceed to the more minute consideration of the intercellular fluid, we must make mention of certain morphological elements which, in addition to the colored cells, are found suspended in the blood: these are the colorless corpuscles to which reference has already been made, and which have been termed *fibrinous flakes*. With regard to the latter, the name indicates that their discoverer, H. Nasse,<sup>1</sup> considers these irregular, crumpled and indented plates, which at most have a diameter of  $\frac{1}{100}$ '''', as a peculiar form of coagulated fibrin,—a view to which Virchow<sup>2</sup> has recently given in his adhesion, but which is opposed by the observations of Henle, Döderlein,<sup>3</sup> and Zimmermann, who found these flakes in uncoagulated blood (both in the fresh fluid and in blood whose coagulation had been impeded by the addition of salts). Hence this substance would necessarily constitute a perfectly distinct variety of fibrin, and therefore a substance which is not fibrin: we have, however, already seen that fibrin itself has never been exhibited in a state of sufficient chemical purity to admit of our calculating a proper formula to represent its composition. But even if we allowed a wide signification to the meaning of the term fibrin, we could hardly regard this substance as fibrin after the chemical reactions which Döderlein observed these flakes to yield; for he found that they were perfectly insoluble in acetic acid (even when its action was much prolonged) and in sulphuric acid, and that they remained for weeks unchanged even after the blood had become putrid. These properties of the flakes are the very reverse of those of fibrin; and to include such a substance under the idea of fibrin, would require a greater elasticity of chemical ideas than is even now allowed. Since the relations of pavement epithelium towards acetic and sulphuric acids and towards putrefaction are precisely the same as those of these flakes, according to Döderlein's experiments, we might assent to the opinion formerly advanced by Henle, and regard the flakes as shreds of epithelium from the lining coat of the vessels, if only there was any coincidence between the forms of the two structures. At present Henle is inclined to regard the flakes as adhering membranes of destroyed blood-corpuscles, to which they certainly bear the most resemblance, as is shown by Virchow's experiments, in which he made the membranes adhere by trituration. In the copiously watered blood of the hepatic veins, which is very rich in these cell-membranes, I also found a large number of perfectly distinct fibrinous flakes, which, like the cell-membranes, were scarcely at all acted on even by acetic acid and alkalies.

I have just read (as these sheets are passing through the press) that

<sup>1</sup> Müller's Archiv. 1841, S. 439, and Handwörterbuch der Physiologie, Bd. 1, S. 108.

<sup>2</sup> Zeitschr. f. rat. Med. Bd. 5, S. 216, and Arch. f. pathol. Anat. Bd. 2, S. 596.

<sup>3</sup> Henle's Handb. d. rat. Pathol. Bd. 2, S. 152.



Bruch<sup>1</sup> believes that he has convinced himself that all the so-called fibrinous flakes are nothing more than *epithelial cells from the skin of the observer*, accidentally falling from the epidermis of the face on the preparation. The occurrence of these fibrinous flakes in most other animal fluids, their absence in the circulating blood, the adhesion of air to them, their chemical relations, the form of the horny epithelial scales, and lastly, the fact that they are found even in a single drop of water over which the head has been shaken, are sufficient grounds for the belief that the majority of the structures which have been regarded as fibrinous flakes are nothing else than dried cells of pavement epithelium; we cannot, however, explain all the formations of this kind, which we sometimes find in the blood, by assuming that they are epidermic scales. If blood has been treated with water (in the same manner in which Nasse treated his fibrin from which he saw such flakes project), we find far more of these fibrinous flakes resembling crumpled laminae than in fresh blood, and this is especially observed when the blood of the hepatic veins is thus treated with water: these are obviously the adhering, stretched, and distorted walls of the blood-corpuscles, which, as we have already indicated, resemble the epidermic scales in resisting the action of acetic acid and of not too concentrated potash lye.

According to the most recent researches, the *colorless corpuscles* are perfectly identical with the lymph- and chyle-corpuscles; indeed, notwithstanding the assertions formerly made to the contrary, no single difference can be pointed out between them and the mucus- and pus-corpuscles: we need only refer to the elaborate works and memoirs of Henle,<sup>2</sup> H. Müller,<sup>3</sup> and Virchow.<sup>4</sup> The corpuscles approximate to the spherical form, and are not elastic; their investing membrane is more or less granular, and is always so viscid that the corpuscles possess a well-marked tendency to conglomerate into larger or smaller groups. In the circulating blood we see them rolling along the walls of the capillaries (while the colored corpuscles move far more rapidly and nearer to the axis of the vessels), as may be easily perceived in the web-membrane of any frog. The contents of the colorless blood-cells consist of an albuminous solution, in which there are suspended extremely fine granules, together with a single, double, triple, or multiple nucleus, which may be either smooth or granular. Water causes the corpuscles to swell, and renders the nucleus visible; the phenomenon is more marked if dilute acetic acid be used, which gradually dissolves the cell-wall, and leaves the nucleus exposed; the endosmotic action of water induces a distinct molecular motion in the granular contents of the cells.

We know far less regarding the chemical nature of the different constituents of the colorless blood-cells, than regarding that of the red corpuscles. As we must notice cells of this kind more fully in our remarks on "Pus," we shall defer for the present any further remarks on what is known on this subject.

There are other morphological elements, as fat-globules, molecular fibrin, &c., which we shall notice when treating of the serum. We make

<sup>1</sup> Zeitschr. f. rat. Med. Bd. 9, S. 216-222.

<sup>2</sup> Allg. Anatomie, S. 422.

<sup>3</sup> Zeitschr. f. rat. Med. Bd. 3, S. 204-268.

<sup>4</sup> Op. cit.

no remarks on the infusoria which some have maintained that they have found in the blood, treating the subject as a long-exploded error.

The textureless fluid constituent of the blood is the intercellular fluid, which, in the circulating blood, contains the fibrin in solution, as well as the constituents of the serum; hence, we first proceed to the consideration of the *fibrin*, and the more so, since from its separation from the blood in the form of the clot, it is closely associated with the blood-corpuscles. As we have already fully noticed the chemical nature of fibrin, we shall here direct our remarks, for the most part, to the mechanical relations which are dependent on the spontaneous separation of the fibrin from freshly drawn blood. We shall therefore, now, principally notice the coagulation of the blood and its results—the clot and its different physical characters.

We have already referred to the views that have been advanced in reference to the cause of the spontaneous coagulation of the fibrin; we have only additionally to mention an hypothesis recently put forth by C. Schmidt,<sup>1</sup> which is essentially very similar to the opinion previously expressed by Schultz. Schmidt believes that the fibrin becomes formed and separates in the following manner: as the blood escapes from the circulation, an acid albuminate of soda which is dissolved in it, becomes disintegrated into its component parts in such a manner, that a less acid, neutral or basic albuminate of soda remains dissolved, while the other atom of albumen separates under the form which we name fibrin; the fibrin subsequently contracts to the smallest possible volume, just as freshly precipitated silica, alumina, and phosphate of lime gradually contract. If we observe the separation of fibrin in threads, &c., it will appear as if the analogy with hydrated alumina, &c., at all events, affords no special support for this hypothesis, which at first sight is sufficiently plausible.

The *coagulation of the blood*—the most striking phenomenon presented by fresh blood—although for a long time the subject of numerous investigations, is still involved in considerable obscurity. We now recognize fibrin as the proximate cause of the formation of the clot; we have also, in the introductory portion of this chapter, explained the process of coagulation, in so far as its external phenomena are manifested in healthy blood: but in various physiological and pathological conditions, we meet with numerous anomalies, whose study promises to elucidate the nature of this process. These anomalies, or rather fluctuations, of the external phenomena, have reference partly to the duration of the individual periods of coagulation, partly to the final consistence of the clot, and partly to the manner in which the blood-corpuscles are enclosed in it. We shall have to seek for the proximate causes of these modifications, partly in the variable quantity and nature of the fibrin, and in the number and character of the blood-corpuscles, and partly also in the chemical constitution of the serum.

We shall first notice the variation in the *time of coagulating*. We much more frequently meet with cases in which the coagulation, or one or other of its stages, is delayed, than in which it is abnormally hastened. In investigating the causes of this difference, we shall at the same time

<sup>1</sup> Charakteristik d. Cholera, u. s. w. S. 205.

become acquainted with the physiological and pathological relations under which the coagulation proceeds either more slowly or more rapidly than usual. H. Nasse must be especially mentioned, as having devoted very great attention to this department of hæmatology, and as having thrown much light upon it by his observations. We must first make mention of certain external relations, which, quite independently of the chemical nature of the blood, exert an influence on the time of coagulation. Among these, we may first notice strong agitation of the blood before, and during the process of coagulation. We find that the separation of the fibrin is more rapidly effected when the blood has been disturbed and shaken, in the same manner as in the case of saturated saline solutions, which deposit their crystals far more rapidly when they have been stirred or agitated. The blood also coagulates rapidly in a vacuum, in consequence of the violent motion induced in the molecules of the blood, by the development of vesicles of gas and aqueous vapor; its coagulation is, however, still more rapid in the air, when the latter is strongly agitated, for here the *access of the air or oxygen* is added to the other accelerating causes. For the same reasons, the rapidity of the coagulation is increased in proportion to the slowness with which the blood flows from the vein, the length of the jet, and the width and shallowness of the vessels in which it falls. Since the blood itself contains gases, the different quantity in which they occur, must necessarily influence the period of coagulation; hence blood which is rich in carbonic acid, coagulates less rapidly than when the contrary is the case; thus, too, the longer retention of the blood in the veins, after the application of the bandage, appears to be connected with an increase of carbonic acid; at all events, we find that the blood coagulates far more slowly than usual when the bandage had been applied a long time before venesection. Moreover, when the exchange of gases is not sufficiently carried on in the lungs, the blood must become poorer in oxygen and consequently richer in carbonic acid; hence the blood coagulates very slowly in cyanosis. A similar reason may also explain, at least in part, the delay frequently noticed in the coagulation of inflammatory blood, and the less rapid coagulation of venous than arterial blood. In prolonged bleeding, the blood that flows last, is found to coagulate much more rapidly than that which escaped first, which is very probably owing to the former containing an increased quantity of oxygen, derived from the deep-drawn and jerking inspirations. This mode of explanation is further confirmed by the lighter color of this blood. The blood taken after death coagulates less rapidly, owing perhaps to its being more abundantly impregnated with carbonic acid.

Another cause which accelerates the coagulation is the *aqueous character of the blood*. According to Nasse's experiments, water accelerates the coagulation of the blood when added in small quantities, or at all events, when not exceeding twice the quantity of the blood, whilst larger quantities tend to retard coagulation. Hence we find that watery blood, as for instance, that of women or after repeated bloodlettings or other losses of the juices, and anæmic blood, generally coagulate more rapidly than blood in a normal state.

It has been long known, that certain *salts*, namely the caustic alkalies

and their carbonates, have the property of retarding, or even wholly arresting, the coagulation of the blood, but the question has not yet been definitely settled in relation to other alkaline salts; for in the experiments on different salts, no attention has been paid to the degree of dilution of the saline solution, or to the quantities of solution employed. Nasse found, however, that almost all salts accelerate coagulation when not employed in too large quantities, although they may retard it when used in very small quantities. On this account, it is far less easy than was formerly supposed to determine the connection existing between the quantity of the salts contained in the blood, and its more or less rapid coagulation in different diseases. Thus the absence of coagulability, which has occasionally been observed in the blood in typhoid and putrid conditions, has been referred to a considerable increase of the salts of the blood, or to the presence of alkaline carbonates, but this is mere opinion, unconfirmed by any experiments. All that can be asserted on this subject, therefore, is that the difference frequently observed in the period in which the blood coagulates in the same form of disease very probably depends upon the amount of salts contained in the blood.

*Viscid solutions* of indifferent organic substances, such as albumen, casein, and sugar, appreciably retard the coagulation of the blood. This circumstance shows us, at all events, how many different conditions may coincide to bring about one or another of these results in reference to coagulation. But here, unfortunately we derive only little aid from chemical analysis; for, as we have already observed, we are still in entire ignorance as to the different quantities of salts occurring in the blood during disease.

The influence of the *temperature* of the blood (as it escapes from the body) on its coagulation, has also been noticed by Nasse, but we are still ignorant how far this may affect the period of the coagulation. The difficulties of investigating more closely the causal connection of the period of coagulation and the external and internal relations of the blood, are further increased by the circumstance, that while these influences are frequently manifested in the blood, they may simultaneously neutralize one another in a greater or lesser degree.

It has likewise been conjectured that the blood when it is rich in fibrin (inflammatory blood) coagulates less rapidly than when it is deficient in that substance; but as the reverse is frequently found to occur, it appears very doubtful whether the quantity of fibrin exerts any influence whatever on the period of coagulation.

In the present state of our knowledge, in reference to the different conditions of the blood, we are alike incapable of explaining why the blood of persons killed by lightning, of those who have died from the effects of narcotic poisons or asphyxia, or from hanging, should not coagulate, whilst it coagulates very rapidly after the infliction of venomous bites, &c., and in the plague.

The *consistence of the clot* is also liable to very great variations. As the *fibrin* actually constitutes the main consolidating substance of the clot, the opinion long prevailed, and has only recently been relinquished, that the cause of this difference was to be sought in a difference in the chemical constitution of this substance; but here we have, in the first

place, to take into account both the external and the internal mechanical influences, which make the clot appear at one time more dense and compact, and at another softer and more gelatinous. The vessel in which the blood coagulates, is not without its influence, for in a shallow vessel, a softer coagulum will be formed than in a high and narrow one.

We reckon, among internal mechanical causes, the relations in which the blood-corpuscles and the water stand to the quantity of the fibrin. When the *number of blood-corpuscles* is small in relation to the quantity of fibrin, its molecules approximate more closely to one another, and the coagulum is more densely compressed. But when an excess of blood-corpuscles is imbedded in fibrin which separates gelatinously, the latter may remain imperfectly contracted during its further consolidation, and thus give rise to a highly friable clot. As the lower part of the clot, moreover, contains the greater number of blood-corpuscles, it is evident that this portion will continue to be softer and looser in texture, whilst the upper part becomes more dense and connected. On this account, we find that the clot in the blood of plethoric persons is large and soft, whilst in that of chlorotic patients it is small and firm.

The fact that too large a quantity of *water* diminishes the consistence of the clot, has chiefly been proved by Nasse, both by direct experiments and by observations on morbid watery blood. It would appear as if the molecules, which are separated in a gelatinous form at the commencement of coagulation, could not be brought into sufficiently close contact with one another to admit of their firm contraction; and hence, the clot may in such cases retain too much serum, which will render it soft and friable. This excess of water may also contribute to produce that greater softness which we observe in the clot of young animals, and may be the cause of the softness noticed in the clot after frequent bloodlettings. As, however, exceptions to these observations sometimes present themselves, we must presume that other influences frequently supervene which counteract the effect of the water. It follows, therefore, that we are unable to draw any conclusions regarding the relations of weight between the serum and the actual coagulum, from the relative volumes of the clot and of the serum; since we should have to consider in such an estimate, whether the fibrin in its condensation had completely pressed out the serum.

Henle further draws attention to a mechanical influence, which may give rise to the formation of a soft and very diffuent coagulum, at least in some few cases; for when the blood slowly flows in *separate drops*, each drop forms, in a certain degree, a coagulum which does not combine with the other drops to form a homogeneous and connected mass. Henle assigns this as the cause of the incoagulable character of the menstrual blood; but Schmidt's and my own observations (to which we shall refer in a future page) have shown that this blood does not contain any fibrin.

The *gases* contained in the blood appear to exert some influence on the consistence of the clot; for whilst a light-red, highly oxygenous blood yields a dense, elastic coagulum, the clot appears to be soft in all conditions in which the blood is rich in carbonic acid; this is especially manifested in asphyxia—a condition in which it has been asserted that the blood exhibits no capacity for coagulation.

It is not impossible that *other constituents of the blood* may influence the consistence of the clot; at all events we find in artificial experiments with salts which retard coagulation, that a soft and frequently even a mere gelatinous coagulum is formed. The soft, friable, and often tar-like consistence of the clot in putrid diseases, may therefore be owing to free alkalies or their carbonates.

We are unable, at the present time, to determine whether the differences manifested in the physical character of the clot, depend upon differences in the *chemical constitution* of the fibrin. Some observers have conjectured that there are different kinds of fibrin; we have already spoken of parafibrin, &c., but no differences in the nature of fibrin admit of being chemically demonstrated; nor are we logically compelled to assume the existence of such differences, since the different forms under which the fibrin coagulates, may possibly depend upon the action of certain chemical relations, of which we are still ignorant. When we remember that ordinary albumen may form either a gelatinous and milky, or a flocculent, or a membranous coagulum, without having experienced any alteration in its elementary composition, we cannot admit the necessity of regarding the buffy fibrin of the inflammatory coat as chemically different from that which is separated, in a crumbling or flocculent form, from tar-like blood.

The *form of the coagulum* mainly depends upon the relations of the blood-corpuscles to which we have already referred. We have seen that, under certain conditions, the blood-corpuscles are disposed to approximate to one another by their flat sides, thus assuming a cylindrical form, and that in this manner they more readily displace the column of fluid which supported them, and sink more rapidly; whilst those cells which are of a jagged, twisted, or spherically distended form, impede such a cohesion, and give rise to a more prolonged suspension of the molecules. The different forms of the clot must, therefore, obviously depend upon the different *sinking capacity of the blood-corpuscles*.

The rapidity or slowness with which the *fibrin* is separated and consolidated, exerts in the same manner a determinate influence on the form of the clot. The differences existing in these proximate causes prove how difficult it is to explain in a special case the remote causes which may give rise to any definite form of the clot. If we here take into account the two proximate causes of difference, namely, the time of coagulation of the fibrin and the sinking capacity of the blood-corpuscles, we find two cases especially which give rise to a different conformation of the clot, namely, *rapid coagulation of the fibrin with little tendency of the corpuscles to cohere*, and *slow coagulation of the fibrin with rapid sinking of the corpuscles*.

Henle first drew attention to the fact that the red sediments of blood-corpuscles, which are often deposited from the blood when there is a dense clot, are owing to the fibrin coagulating and becoming contracted before the corpuscles had assumed the roll-like or nummular form; on the contraction of the gelatinized fibrin, a large number of the loosely connected or wholly isolated blood-corpuscles are again expressed, by which means the serum is for a time rendered turbid and red, until they afterwards separate into the above-mentioned sediment, which readily

admits of being again disturbed. Zimmermann, who confirmed this view of Henle's by a microscopical examination of the red deposit, found, moreover, that besides this sediment there was always present a small but very compact clot, which proves that the coagulation of the fibrin acted an important part in this phenomenon.

We far more frequently observe the converse relations in diseased, and even sometimes in healthy blood—that is to say, the corpuscles have a strongly marked sinking capacity, whilst the fibrin coagulates slowly. We must here bear in mind that extreme cases are of the rarest occurrence, and that both properties are wholly relative; for in the one case the fibrin may contract as usual within an average time of coagulation, while the corpuscles sink more rapidly; and in the other case, the corpuscles may sink with their ordinary velocity only, while the fibrin, on the contrary, coagulates very slowly. The result will be much the same in both cases. The influence of these two causes may be perceived even in the normal clot, for here we find that the lower part of the clot is always darker and softer than the upper one; this depends, certainly, only in part upon the circumstance that there are more blood-corpuscles which have already sunk in the lower than in the upper portion, for the light color of the upper part depends on the one hand upon the access of oxygen, and on the other upon the larger number of colorless blood-corpuscles, which, although they combine in groups, owing to their viscosity, are not so very closely in contact in consequence of their spherical forms, and do not, from their lightness, sink as rapidly as the red corpuscles. When the red corpuscles of fresh blood have sunk in some degree before the fibrin becomes gelatinized, the fibrin coagulating in the uppermost stratum of fluid is unable to enclose any red corpuscles, and consequently forms a colorless crust upon the subsequently deposited clot. As this crust encloses only few foreign elements, the fibrin of which it consists contracts more closely than that which is beneath it, and in which the blood-corpuscles are embedded. This crust will therefore not only present a smaller diameter than the red clot, but it must also, from its contiguity, cause an extension of the margins of the latter, while it gives rise to a concavity of the clot. This concave and generally very compact and yellowish-white *buffy coat* is of most common occurrence. It is principally found to occur in the venous blood of horses and in inflamed blood, and sometimes also in human blood, if drawn during the process of digestion. A plane or convex buffy coat is also observed in many morbid conditions; in these cases it is soft, and of a grayish-white color; and it is not improbable that this character depends no less upon an excess of colorless blood-cells and vesicles of fat in the crust, than upon the slight contractility of the fibrin.

Although this mode of explaining the formation of a fibrinous or buffy coat scarcely needs any additional grounds for its establishment, Müller, H. Nasse, and Henle have confirmed it by special experiments; for they formed an artificial buffy coat from blood that was not buffed, by employing means either for accelerating the sinking of the blood-cells, or for retarding the coagulation of the fibrin. Nasse found, moreover, on comparing the blood of different animals, and by closely examining diseased buffy blood, that the time in which the blood-cor-

puscles sink bears an inverse relation to that in which the fibrin coagulates. Nasse and others have, however, also frequently seen cases in which rapidly coagulating blood formed a buffy coat; these instances do not, however, present any exception to the rule, since they only show that the sinking of the corpuscles has proceeded more rapidly than the coagulation of the fibrin.

We must not omit all mention of certain relations which, although they do not constitute the sole conditions necessary for the formation of a buffy coat, may contribute simultaneously with other causes toward its production. Among these we must first instance *the form of the vessel* in which the blood is suffered to coagulate. In a high narrow vessel, the blood-corpuscles are sooner removed from the level of the fluid than in a wide and shallow one, and thus leave a part of the fibrin to coagulate without them; on this account, strongly inflamed blood is often found to yield no buffy coat in a flat vessel, whilst, on the contrary, blood which is considered to be of a non-inflammatory character exhibits a buffy coat if received in a narrow cylinder.

The *number of the corpuscles* is another cause which contributes to the formation of this coat. When the number of corpuscles is small and their sinking capacity is considerable, a buffy coat will more readily be formed than where the blood-corpuscles are present in large numbers. On this account a buffy coat is more frequently formed after a second or third, than after the first venesection. The same cause explains its more frequent occurrence in the blood of anæmic and pregnant females, than in that of healthy and non-pregnant women.

It was formerly regarded as a well-established view, that the formation of this coat was owing to an *excess of fibrin*; and as the increase of the fibrin was in general proportional to the progress of inflammation, it was also termed the inflammatory crust. It cannot be denied that the quantity of the fibrin exerts some influence on the thickness of the buffy coat, but it can never constitute the sole cause; for we frequently observe inflammatory blood, which is very rich in fibrin, form no crust, whilst, as we have already noticed, blood that is poor in fibrin may in many chronic affections present a crust of this nature.

We have, therefore, to revert to many different proximate or remote mechanical and chemical causes for an explanation of the configuration of the clot in any special case. The observations regarding the mechanical relations of the clot are only important in a semeiotic point of view in special cases; they are of no service in the establishment of artificial families or groups of diseases.

Before we pass to the further consideration of the substances actually dissolved in the intercellular fluid, we have to notice some bodies which remain suspended in the blood, or more correctly speaking, in the *serum*, after the separation of the fibrin and the blood-corpuscles.

Hewson and Thomson are of opinion that they have found a milky *turbidity of the serum* in blood taken some hours after a meal; but I have never observed anything of this kind either in carnivorous or herbivorous animals, whose blood I have examined at different periods after the administration of food. In a similar manner the serum acquires, according to Hewson and Magendie, a milky appearance after prolonged



fasting. Nasse found that the serum of the blood of pregnant women was in most cases milky; very frequently, but not invariably we observe that in drunkards the serum presents an opalescent, almost milky turbidity. This turbid appearance is generally due to the presence of suspended *fat*, which may easily be detected by microscopical examination or by shaking the serum with ether.

Zimmermann has drawn attention to a kind of turbid serum, which he has found in the blood in inflammatory conditions. This turbidity depends upon the presence of very small dark particles, or molecular granules; and hence he was induced to assume the existence of a special molecular fibrin, while Scherer, on the contrary, who has noticed similar instances of turbidity, inclines rather to the opinion that these granules are separated albumen. My own observations on the turbidity arising from molecular granules lead me to concur in the latter view; for I found that the turbidity disappeared on the addition of neutral alkaline salts, when the serum exhibited only a faint alkaline reaction; and hence we must assume, with Scherer, that a portion of the alkali is by some means removed from the albuminate of soda in the blood, and that a portion of the *albumen* has been separated in a finely granular form, after the removal of the alkali which had been combined with it (sec p. 297).

In some cases the turbidity of the serum depends upon the presence of suspended *colorless blood-cells*, as both Pieschel and myself have observed in the blood of dogs affected with the mange.

If it be admitted that the physical relations of morbid blood, as it flows fresh from the vein, is a subject of considerable importance, the character of the *blood in the dead body*, in respect to the mode of its coagulation and its color and consistence, cannot be devoid of interest to the pathologist. However strongly we may protest against the doctrine of crases, which is based on such investigations, as a perversion of the so-called pathologico-anatomical tendency, we cannot withhold our testimony to the value of the labors of such inquirers as Rokitansky and Engel. The connections which Engel has ingeniously established between the nature of the blood in the dead body and its imbibition in the tissues, its accumulation in separate organs, and the character it impresses on the individual tissues, as well as regarding the nature and extent of the preceding exudations or transudations, show that we are justified in looking for a rich accession to our scientific knowledge from a more exact chemical investigation of this subject. Unfortunately, however, no chemist has as yet been inclined to direct his attention to this subject. Hence we do not deem it altogether superfluous to follow the arrangement of Rokitansky and Engel, and to consider the different kinds of blood in the dead body with reference to the above physical properties, classifying them in the six following groups:—

1. One kind of blood found in the dead body is distinguished by its thick fluid character, its reddish-brown color, and its coagulability; and is probably for the most part characteristic of a certain group of diseases, since it is only met with in the bodies of persons who have died from violent inflammations (with the exception of inflammatory affections of the brain, and the spinal cord). Blood of this kind becomes bright red on exposure to the air, and coagulates only in the larger vessels; in the

capillaries and the smaller vessels it retains its thin fluid nature, and the coagula which occur in the heart and in the larger arteries, as well as the larger veins, are almost always compact, and of a dark brownish-red color. The thickness of this blood is the cause why it is less readily infiltrated into the tissues than other blood. It is moreover worthy of notice, that fibrinous coagula never occur simultaneously with these clots in the heart and the larger vessels, for when they are present, they are found in the vessels of moderate calibre, but never in the capillaries.

2. The blood in acute diseases of the spinal cord and of the brain is found to be thickly fluid, of a dirty brownish-red color, uncoagulated, and devoid of fibrinous coagula.

3. A thick, uncoagulated, and not coagulable blue and blackish-red blood, which, under certain favorable conditions, sometimes deposits fibrinous coagula in the heart and in the larger vessels, is certainly not characteristic of merely one form of admixture of the blood; for blood of this kind is found in the body after diseases which reciprocally exclude one another, as, for instance, after plethora (depending upon heart diseases), typhus, acute tuberculosis, cholera, and poisonings with narcotics and lead, and after sudden and profuse sweats or diarrhoeas.

4. A pale, or vermilion-red, uncoagulable, thin, fluid blood, which, notwithstanding its fluidity, does not readily infiltrate the tissues, but which often deposits a considerable quantity of fibrinous coagula in the larger vessels, does not belong to any special admixture of the blood, since it is met with after the most varied conditions of disease, when the blood has acquired a watery character; as, for instance, after frequent venesections, hemorrhages, considerable exudations, long-continued diarrhoea and sweats; and in the anæmia following typhus and the acute exanthemata, as well as in senile atrophy.

5. A thin bluish-black, uncoagulable blood, which is distributed in large quantities from the great to the smallest vessels, which easily infiltrates into the different tissues, and which never exhibits any separation of fibrinous coagula, is found in valvular anomalies of the heart.

An accurate analysis of this variety of blood, compared with the composition of the blood in the living body, during the existence of the different conditions arising from mechanical difficulty and obstruction of the function of respiration, as, for instance, plethora, hemorrhoidal affections, dropsy, &c., would undoubtedly yield the most valuable aid towards the explanation of the mechanical and chemical metamorphosis of matter in the animal body.

6. Finally, there is a kind of blood found in the body after death, which is thinly fluid, uncoagulable, and of a dirty brownish color, does not deposit fibrinous coagula, is easily infiltrated into the tissues, but is generally found to occur in inconsiderable quantities in the heart and the large vessels, while it is accumulated in abundance in the capillaries. This condition is observed in true decompositions of the blood, as for instance, in pyæmia, puerperal fever, scurvy, &c.

We are still in the dark as to the direct origin of those polypous coagula of fibrin which are deposited from uncoagulable blood containing but little fibrin; and all that we know in reference to the subject is,

that the retarded circulation induced shortly before death by mechanical obstruction as well as by debility, is favorable to the deposition of these masses—and hence their occurrence after a protracted death-struggle. The formation of the purely local fibrinous coagula observed in aneurisms, obliteration of the veins, phlebitis, &c., may be explained in a similar manner.

We now pass to the consideration of the actually *dissolved chemical constituents of the serum*; amongst which we must first direct our attention to *albumen*. As we have already fully considered this substance in its various relations to other protein-bodies, and to the other constituents of the blood, it only remains for us to notice one or two additional points.

The question has often been raised, whether the albumen in different vessels, under different physiological relations, and in different pathological conditions, is always identical. Physiological, no less than logical grounds, warrant us in answering this question in the negative, although chemistry affords but little aid in determining the point. We have already seen (p. 296) that many modifications in the properties of albumen depend upon the different quantity of alkali or salts which it contains, while the organic group of atoms in the albumen has always remained the same. Differences in the albumen, depending upon an augmentation or diminution of the quantity of alkali, that is to say, neutral, basic, and acid albuminates of soda, occur even in the normal condition, as for instance, in the blood of different vessels. The solution of neutral albuminate of soda becomes turbid on the addition of water. This combination occurs not only in morbid blood (as Scherer was the first to show), but also in the blood of different vessels, as also in the blood of the splenic vein; and here, independently of the other metamorphoses experienced by the blood in the spleen, a portion of the basic albuminate of soda is saturated by the free acid which we find in the parenchyma of that organ, and the neutral compound is thus formed. The serum of the portal blood appears, moreover, less turbid on the addition of water than that of the splenic vein; while that of the hepatic veins exhibits great turbidity on the addition of water; and here the albumen of the portal vein, probably, loses a portion of the alkali which is applied to the formation of the bile.

It appears from the investigations of Guillot and Leblanc,<sup>1</sup> Panum,<sup>2</sup> Stas,<sup>3</sup> and others, that the substance which they regard as *casein*, is contained in a larger quantity in the blood of pregnant and puerperal women than in ordinary blood. The supposed occurrence of casein in the blood has been already noticed.

Notwithstanding the proofs which Panum and Moleschott have brought forward to demonstrate the existence of casein in the blood, Scherer is still by no means convinced that their casein is anything more than albuminate of potash or soda. See p. 297.

In accordance with Scherer's view, we must regard the different form in which the albumen coagulates, as depending upon the different quantity of alkali which it contains (see p. 296); but still we often observe, that where the alkaline fluid has been neutralized or faintly acidified, in order

<sup>1</sup> Compt. rend. T. 31, p. 585.

<sup>2</sup> Arch. f. path. Anat. Bd. 3, 268.

<sup>3</sup> Compt. rend. T. 41, p. 629.

to induce such a complete deposition of the albumen as may be necessary for its perfect filtration, the albumen of one blood collects less easily in flakes, and is less readily filtered, than that of another. Thus, I constantly found that the albumen of the blood of the hepatic veins only accumulated in masses very slowly, often not till it had been boiled for hours, whilst that of the portal and other veins, as well as that of the arteries, very readily coagulated on boiling after the addition of an acid, and that it rapidly sank, leaving a clear supernatant fluid.

Since, as we have already observed, ordinary chemical investigations, and more especially elementary analyses, still fail to throw much light on the inquiry into the essential differences in the protein-bodies, C. Schmidt<sup>1</sup> has conceived the happy idea of bringing substances that are readily capable of fermentation or decomposition into contact with the constituents of the blood under favoring conditions, and thus employing sugar, urea, amygdalin, asparagin, &c., as tests for the presence of certain modifications of albumen; as yet, however, the only results at which he has arrived, are that the blood-cells of a healthy individual (but not the intercellular fluid) contain one substance which yields sugar-ferment as the product of its spontaneous decomposition, and another which similarly yields urea-ferment. In diseases, as for instance in cholera, one or the other of these fermenting bodies is increased to a great degree.

We have already fully considered the fats of the serum, and we need, therefore, here simply remark that only a small quantity of free fat occurs in the serum, while saponified fat is always present in large quantities, as well as the crystallizable lipoids, cholesterin, and serolin. It cannot be shown, with certainty, that the serum contains any phosphorized fats. We shall perceive from special numerical results, that the quantity of the fat, and also the kinds of fat occurring in the different veins and under different physiological relations, present great variety. The fat of the serum as compared with that of the blood-corpuscles, may be regarded as more readily crystallizable, and less tenacious and colorless, but far inferior in respect to quantity. The difference between the quantity of fat contained in the intercellular fluid and that of the blood-corpuscles is obvious from the above review of the quantitative distribution of the different constituents in the corpuscles and the intercellular fluid. We would only observe, therefore, that the considerable quantity of fat which is mixed with the fibrin, has very frequently been regarded as peculiar to that constituent. Virchow<sup>2</sup> found from 2.50 to 2.76% of fat which could be extracted with alcohol and ether in human venous fibrin, Schmid<sup>3</sup> from 4.21 to 5.04% in the fibrin of the jugular vein of horses, and from 7.37 to 8.72% in that of the portal vein, whilst I found 2.154 in the buffy coat of the venous blood of the horse, and 2.168% in the arterial blood of the same animal.

It is certainly of some importance in a physiological point of view to decide whether the fat which can be extracted from this substance is of a peculiar kind, and exists in chemical combination with it, or whether it is only incidentally mixed with this substance, that is to say, from

<sup>1</sup> Charakteristik der Cholera, S. 57-68.

<sup>2</sup> Zeitschr. f. rat. Med. Bd. 4, S. 266-293.

<sup>3</sup> Heller's Arch. Bd. 4, S. 322.

purely mechanical causes. It has been usual to follow the views of Berzelius in this respect, and to regard this fat as peculiar to the fibrin, and as distinguished from other fats of the blood by the amount of nitrogen it contains; but a more attentive consideration of the mode in which the fibrin is exhibited, leads us to doubt the correctness of this opinion. We have here only to consider the mode of preparation of the fibrin, and the admixtures which it always contains; the fibrin in its spontaneous coagulation must necessarily draw down and enclose particles only suspended in the blood, and in addition to these and the blood-corpuscles, occasionally very minute fat-vesicles, and always colorless blood-cells. When the fibrin is obtained by washing the clot, the granular contents of many colored blood-cells, which mainly consist of fat, remain in the fibrin together with the cell-walls. We have already shown in an earlier part of this work that many colorless blood-cells are mixed with the fibrin, and it must further be observed, that they contain, absolutely and relatively, more fat than the colored cells. We do not call in question the possibility that acid salts of the fatty acids in the serum (enclosed in the clot) may be rendered insoluble by strong dilution with water. There are at all events, a number of possible sources to which the fibrin-fat may and indeed must in part be referred. It is only necessary, therefore, for the determination of this question, to ascertain whether this fibrin-fat differs specifically from the fats associated with the other constituents of the blood. Such, however, does not seem to be the case, for as far as my own and Virchow's inquiries extend, the fibrin only contains fats which belong to some one or other of the blood-constituents. Virchow found a considerable quantity of acid phosphate of lime in the ash of this fat; the other reactions of the fat seemed also to confirm the presence of glycerophosphate of lime, which, as we have seen, is peculiar to the colored blood-cells. There is also an acid ammonia-soap contained in the fibrin-fat, which may possibly have been conveyed to it by the serum. We know so little of the non-saponifiable fats, that they cannot aid us in deciding this question either negatively or affirmatively. Virchow could not detect cholesterin in the fibrin of man, but I have demonstrated its presence in the fibrin of horses by the micrometrical measurements of its angles. It might indeed also be derived from the serum. It follows from the above observations, that we are not as yet justified in ascribing special fats to the fibrin. We might perhaps be disposed to attach some weight, as far as this question is concerned, to Virchow's observation of an acid reaction of the fat of the fibrin, but independently of the fact that the fat of the colored blood-cells exhibits a similar reaction, there are several grounds for explaining this phenomenon. In the first place, these fats, as they contain the salts of the fatty acids, must assume an acid reaction whenever the ether which is employed, is not entirely pure (free from acetic acid, aldehydic acid, &c.), and from several investigations in relation to this subject, I am disposed to think, that the metamorphosis of the ether into acids, by the action of animal substances, is considerably promoted by prolonged digestion. On the other hand, we can the less wonder at the acid reaction of these fats, since the salts of the fatty acids precipitated with the fibrin by water, are acid salts, from

which on fusion volatile and acidly reacting fatty acids are separated from their combinations with bases. Thus, for instance, we constantly find volatile fatty acids in these fats (both in those of the fibrin and of the blood-corpuscles), as for instance, acetic acid, which may be produced by the metamorphosis of the ether, and at least *one* acid, which, when treated with baryta, yields a salt which crystallizes into beautiful laminæ, and which undoubtedly belongs to the group of the true volatile fatty acids.

With regard to the *extractive matters* of the serum, it is better to pass them over in perfect silence, than to collect the fragmentary and inconclusive facts regarding them at present in our possession. From the physiology of the metamorphoses going on in the blood, we should be led to suppose that the extractive matters are far more abundant in the intercellular fluid than in the blood-cells, and this view is confirmed by direct experiment; for, as is obvious from what has been already stated regarding the composition of the blood, these substances occur more abundantly, both relatively and absolutely, in the serum than in the cells.

A number of substances were formerly included and concealed among these extractive matters of the blood, and especially of the serum, which have either lately been discovered or which we have not yet succeeded in finding. First amongst these we must place *sugar*. This has very recently been proved by C. Schmidt<sup>1</sup> to be an integral constituent of the normal blood of cattle, dogs, cats, and diseased and healthy men. It has been already mentioned, that in consequence of Bernard's discovery of sugar in the liver, I sought for this substance in the blood of the portal and hepatic veins, and found 10 or 12 times as much in the latter as in the former, in which the quantity was very small.

The method of determining the amount of sugar in the blood is very simple: freshly drawn and defibrinated blood is gradually treated with 8 or 10 times its volume of alcohol, the mixture being thoroughly shaken; the coagulum is washed with hot alcohol, and the alcohol of the filtered fluid driven off; the residue is further concentrated, and then extracted with stronger alcohol, whereby the greater portion of the salts is separated; a part of the alcoholic fluid is now treated with an alcoholic solution of potash, by which the potash-and-sugar compound and a little extractive matter are precipitated; the precipitated flakes become caked together on the filter, from the action of the air; we then dissolve them in water, and can determine the sugar qualitatively by Trommer's, and quantitatively by Fehling's test. Another part of the alcoholic solution (from which the greater part of the salts has been removed in the above-described manner) may be evaporated, dissolved in water, and treated with a little yeast; and the quantity of sugar can then be calculated from the carbonic acid which is evolved. (See page 253.)

Other substances occurring in the serum of normal blood are *urea*, *uric acid*, and *hippuric acid*. The latter has been found by Verdeil and Dollfuss<sup>2</sup> at Giessen, in the blood of oxen. That *creatine* and *creatinine* occur in the blood, has certainly not yet been proved by

<sup>1</sup> Charakteristik der Cholera, u. s. w. S. 161-164.

<sup>2</sup> Compt. rend. T. 30, p. 510 et 657-660.

direct investigation, but from the simultaneous occurrence of these two substances in the muscular juice and in the urine, we are justified in concluding that they exist there.

Whether *biliary matters*, to wit, the *biliary acids*, occur preformed in normal blood, we have not at present the means of deciding (as has been already mentioned); on theoretical grounds, we should, however, regard their presence as improbable.

We are still perfectly in the dark regarding the *pigments* of normal serum.

The faint yellow color which is peculiar to normal serum, certainly does not depend on bile-pigment; at all events, we cannot exhibit the well-known and striking reactions of cholepyrrhin with the extracts of the serum. In diseases, the serum often assumes an intense yellow color, with or without simultaneous turbidity; this depends either on bile-pigment, which is recognizable in the blood, not merely in icterus, but sometimes also in cases of pneumonia, or on an augmentation of the above-mentioned little understood serum-pigment (which is also most frequently observable in inflammatory processes), or lastly, on suspended blood-corpuscles. Schultz is of opinion, that hæmatin may also occur in solution in the serum, if the contents of the blood-cells become diffused in it, in consequence of a deficiency of salts in the blood. Such cases must, however, be very rare.

We have little to add to what has been already stated, regarding the *salts* peculiar to the serum. While phosphates and potash-salts predominate in the blood-corpuscles, we find a preponderating quantity of *soda-salts*, and especially of the *chloride of sodium*, in the serum; on an average we also find far more salts in the serum than in the blood-cells (after deducting the iron). *Alkaline sulphates* and *carbonates* belong also principally to the intercellular fluid.

Before concluding our remarks on the qualitative examination of the blood, we must mention the persistent *odor* which is peculiar to that fluid, and which is particularly evolved on mixing blood with a larger quantity of sulphuric acid, as for instance, 1 volume of blood with  $1\frac{1}{2}$  of acid. Barruel<sup>1</sup> believed that he had ascertained that the blood of every animal possesses its own peculiar odorous principle, and stands in a definite relation with the odor of the cutaneous and pulmonary transpiration. These and several other opinions of Barruel, having reference to medico-legal investigation, have not been altogether confirmed. They have been submitted to a very careful experimental criticism by Schmidt,<sup>2</sup> who found that the peculiar odor was evolved in an unmistakeable manner by the blood of the goat, the sheep, and the cat, while the odor which was developed from the blood of other animals did not possess a distinctly specific character.

According to Barruel, the odorous principle of the blood is more distinct in the male than in the female sex in every species of animal; as, moreover, it may be developed from the serum, it would appear to pertain to that portion of the blood. Further, the manner in which this odor is developed, indicates that we are here dealing with volatile acids

<sup>1</sup> Ann. d'Hygiène publique. N<sup>o</sup>. 6. 1829.

<sup>2</sup> Diagnostik verdächtiger Flecke in Criminalfällen. Mitau u. Leipzig, 1848, S. 19.

which belong, or at all events are closely allied to the butyric acid group.

The general remarks which we have made regarding the analysis of the animal fluids, especially apply to the *analysis of the blood*. The shortest possible critical review of the different modes that have been adopted for analyzing the blood, will fully confirm the truth of those observations.

One of the most important deficiencies in the analysis is obviously connected with the circumstance that the primary and most important physiological question, namely, the quantitative relation between the fresh *blood-corpuscles* (with their moist contents) and the plasma belonging to them, cannot be answered in the present state of analytical chemistry. We must hence rest satisfied with determining, at all events approximatively, the solid, coagulable and insoluble constituents of the blood-corpuscles; we say approximately, for even the methods of determining the insoluble matters of the blood-cells have in part only a relative value; their quantity is usually not directly found, but calculated from several determinations; moreover, the originator of every indirect method of determining the blood-corpuscles must admit that this method never can give a perfectly correct result, even for hypothetically dry blood-cells, since it is impossible to declare, by any of these indirect methods, how much of the constituents of the serum enclosed in the clot, is still adhering to the blood-corpuscles, and must accordingly be deducted. The worst deficiency in all blood-analyses however is, that the errors are not constant, even when the same method is employed; that is to say, the acknowledged error in every analytical method is of variable magnitude, so that even the comparative blood-analyses made according to one and the same method—a procedure on which the French chemists lay such stress—have only a very subordinate value for physiology and pathology, and the conclusions drawn from them can only be received with the most extreme caution. We must confess with sorrow, that even at the present day the analysis of the blood must be ranked amongst the most uncertain and untrustworthy investigations in the whole department of analytical chemistry. Hence, the attempt which has been recently made (by Hinterberger under the superintendence of v. Gorup-Besanez<sup>1</sup>) to prove experimentally the comparative certainty of the different methods of analyzing the blood, is the more praiseworthy; it is only in this way that we shall attain to what at present seems in some measure impossible. We ought not, however, to expect that chemistry at its beginning should equally distribute its full light over a field on which scarcely a glimmer of twilight has fallen during preceding centuries of investigation.

Most of the experimenters who have made large series of blood-analyses, namely, Andral and Gavarret,<sup>2</sup> Becquerel and Rodier,<sup>3</sup> and Popp,<sup>4</sup> have scarcely at all deviated from the method by which Prevost

<sup>1</sup> Arch. f. phys. Heilk. Bd. 8, S. 603–618.

<sup>2</sup> Ann. de Chim. et de Phys. T. 55, p. 227.

<sup>3</sup> Gaz. Méd. de Paris, 1844. No. 47, p. 751.

<sup>4</sup> Untersuchungen über die Beschaffenheit des menschl. Blutes in verschiedenen Krankheiten. Leipzig, 1845, S. 68.



and Dumas<sup>5</sup> determined the dry blood-corpuscles. This method consisted, essentially, in separately weighing the serum and the clot, after the perfect contraction of the latter, in order to determine the ratio in which they stood to each other; the solid residue of the serum was then determined, as also was that of the clot; on deducting the fibrin, which had been otherwise determined, from the solid residue of the clot, we obtain the number which expresses the sum of the dry blood-corpuscles and of the solid residue of the serum still enclosed in the clot. It is in the accurate determination of the amount of this serum that our most able experimentalists have broken down. Since the amount of water in the clot probably stands in a near ratio to that of the serum, Prevost and Dumas unquestionably believed that they were most nearly approximating to the true ratio, when they regarded all the water found in the clot as pertaining to the serum, and calculated accordingly the amount of the solid constituents of the serum contained in the dry clot, the amount of fibrin being previously deducted.

Since the whole of this calculation depends on a mere simple proportion, it would be quite superfluous to enter into further particulars regarding it.

I cannot imagine, as C. Schmidt appears to assume, that Prevost and Dumas actually believed that all the water of the clot depended only on the serum, but I think it more than probable that they took the view which we have already described. Since the quantity of the serum enclosed in the clot could not be absolutely determined, and as there was no available means of estimating it, the only alternatives that remained for them were, either to calculate all the water of the clot (after deducting the fibrin) as belonging to the blood-corpuscles alone, or as belonging to the serum alone; and they chose the latter. Neither they nor any of their followers have supposed that either of these views was not decidedly erroneous; they, as a matter of course, chose that which obviously led to the smaller error. Von Bibra seems, therefore, to have fallen into a mistake, in believing that by disregarding the serum contained in the clot, he could diminish the error of these chemists.

The modifications of this method which other experimentalists, namely Becquerel and Rodier, and Popp, have adopted, all retain the same error to which we directed attention in speaking of the plan originally made use of by Prevost and Dumas; the former determined the solid residue of the defibrinated blood, and deducted from this the solid residue of the serum which they calculated from the amount of water (the solid residue being determined from a separate analysis of the serum). Popp analyzed the serum that separated from defibrinated blood, and then the cruor which was formed under this serum. These modifications, although not essential, are undoubted improvements on the original method; for it would be folly to attempt the impossibility of thoroughly drying the clot, as it is obtained from the coagulation of the blood; if, however, we take a portion of the clot for the determination of the solid residue, we must at all events adopt the caution of analyzing a vertical section of it, since the corpuscles are very unequally distributed in the clot

<sup>5</sup> Ann. de Chim. et de Phys. T. 23, pp. 56-75.

from above downwards. In many cases it is better to separate the serum from the cruor, according to Popp's method, than from the clot according to the method of the French chemists. This separation of the serum from the sunk corpuscles by any method is, however, in most cases, the most uncertain part of the analysis; for in drawing or pouring off the fluid portion from the clot, it rarely happens that the serum is obtained perfectly free from blood-corpuscles, or the clot perfectly free from serum, independently of the quantity which is enclosed.

Although, however, all authors have been compelled to admit that this mode of determining the dry corpuscles can have no absolute value, it has been generally regarded as perfectly available and sufficient for comparative analyses of the blood; but we must remember with what very different power the fibrin contracts in the clot in different diseases; a very dense clot will enclose far less serum than a very loose gelatinous one; and we do not take into account, that sediments of blood-corpuscles often occur external to the clot: further, the serum obtained from the blood-corpuscles occurs in no definite proportion, since the quantity of serum remaining mixed with the corpuscles is less dependent on the manipulation of the operator than on accident.

Simon<sup>1</sup> struck upon a method of finding the quantity of the blood-corpuscles directly, which however is altogether wanting in accuracy. He coagulated whipped blood by the application of heat, stirring or shaking it the whole time, and then extracting the coagulum with ether and boiling alcohol: he thought that boiling alcohol left the albumen of the serum in a state of purity, and dissolved the constituents of the blood-corpuscles together with the salts and extractive matters of the serum; after the evaporation of the alcoholic solutions, the residue was extracted with cold aqueous spirit, which, as Simon appeared to believe, left undissolved all the constituents of the blood-corpuscles, while it dissolved the non-coagulable matters of the serum. This method presents so many imperfections that one only wonders how Simon's blood-analyses should coincide so tolerably well with those of other experimenters. In illustration of the utter unfitness of this method it may suffice to mention, that two analyses of one and the same blood, made according to Simon's directions, would never by any chance coincide. This method, in consequence of its minuteness of detail, has never been adopted in large series of blood-analyses.

Scherer<sup>2</sup> has in many respects improved the analysis of the blood, and his method is the most correct that has yet been suggested, although it presents the same prominent deficiency as the others, namely, the mere determination of those constituents of the blood-corpuscles which are coagulable and insoluble in water, together with the uncertainty attaching to the absolute value in consequence of the impossibility of estimating the serum that is actually enclosed. Thus, Scherer does not compare the solid residues of the serum and the defibrinated blood, but the quantities of the coagulable constituents of both fluids, in order to find the number of dry blood-corpuscles, and calculates the salts, fats, and extractive matters independently. From the compa-

<sup>1</sup> Med. Chem. Bd. 2, S. 83 [or English Translation, vol. 1, p. 175].

<sup>2</sup> Otto's Beitrag z. d. Analysen gesunden Bluts. Würzburg, 1848.

rative investigations of Hinterberger, it appears that Scherer's method yields the smallest number for the blood-corpuscles, and the reason of this is easily seen; for the dry corpuscles, in Scherer's analyses are deprived not only of all their soluble constituents, but also of an undetermined quantity of earthy phosphates, by the acetic acid employed in the coagulation, and in addition to this, a little pigment sometimes remains in solution, in the fluid, notwithstanding the boiling and neutralization, and there is then so much lost in the calculation of the blood-corpuscles. The principal reason may, after all, lie, as v. Gorup-Besanez and Hinterberger have suggested, in the manner in which Scherer obtains the defibrinated blood, which is as follows: he applies pressure to the clot, and mixes the fluid which escapes with the serum—a method of procedure by which a greater or lesser number of corpuscles, or at all events of their remains, must invariably be retained in the fibrin, and thus be lost in the determination of the mass of the dry blood-cells.

We now arrive at a method which appears to avoid the errors of those we have previously described, and to separate the whole of the serum from the corpuscles. It is based on the property (mentioned in p. 564) which a solution of Glauber's salts possesses of rendering the blood-corpuscles capable of being retained on a filter. It was first applied by Figuier, and subsequently improved by Dumas, and more recently by Höfle.<sup>1</sup> Defibrinated blood is treated with eight times its volume of a concentrated solution of Glauber's salts and filtered, the residue on the filter is rinsed with the same solution (Dumas simultaneously conducts a stream of oxygen through the mass lying on the filter), and finally the mass of blood-cells retained on the filter is either directly coagulated with hot water (Figuier), or is first washed off the filter in tepid water, and then coagulated by boiling. Practicable and accurate as this proceeding appears at first sight to be in theory, it is not to be depended upon in practice. Notwithstanding the precautionary rules recommended by Dumas, some of the blood-corpuscles almost always pass through the filter, and this is the more likely to occur, the more rapidly the corpuscles adhere in dark-red masses to it; but even when the filtered fluid appears only a little colored, we can always detect plenty of corpuscles in it with the microscope, or at all events perceive the deposition of a red sediment; the fluid often passes so slowly through the filter, that the latter becomes completely blocked up by the more or less altered blood-corpuscles. This method of procedure is very often inapplicable to diseased blood, either from its corpuscles passing as readily through the filter after the addition of sulphate of soda as before (Didiot and Dujardin<sup>2</sup>), or because the serum is so viscid and almost gelatinous, that it will not pass through the filter. In a very small number of cases this difficulty may be got over by substituting a solution of sugar for one of sulphate of soda (Poggiale<sup>3</sup>). This is, however, the most important question—Is all the serum actually separated in this manner? If this were the case, this method might at all events be used as a check to other methods, as for instance, to Scherer's, and we should thus probably be able to discover a coefficient for the error (consequent on the serum

<sup>1</sup> *Chemie u. Mikrosk. am Krankenbette.* S. 132.

<sup>2</sup> *Comp. rend. T. 23, p. 227.*

<sup>3</sup> *Ibid. T. 25, pp. 198-201.*

retained in the clot) which is unavoidable in the preceding methods; but unfortunately this is not the case, for the corpuscles collected on the filter are by no means free from serum after two or three washings with a solution of Glauber's salts, as Höfle believes; for the fluid running off the corpuscles even after six or eight washings, is not free from the constituents of the serum (if indeed so many washings do not cause the disintegration of the corpuscles or the clogging up of the filter); this is the reason why, as Gorup and Hinterberger found, this method yields more dry blood-cells than any other, notwithstanding the above-mentioned loss of corpuscles and their constituents (which, when the globulin of the blood-cells is imperfectly coagulated, remain dissolved with the sulphate of soda, especially if a little acid has been added to the fluid to be coagulated). This excess of blood-cells becomes more intelligible when we have convinced ourselves (as I have often done) that clear blood-serum becomes strongly turbid by a saturated solution of pure sulphate of soda. Hence, notwithstanding the most careful washing, substances are added by the serum to the corpuscles. Moreover, in examining the ash of the blood-cells determined by Höfle's method, Hinterberger found a large amount of sulphates. This, however, I have never observed when the coagulum was properly washed with hot water. But, on carefully considering the application of this method, we find that in theory also there are certain objections to it. Thus, if we wash the blood-corpuscles with a fluid which leaves the walls of the cells uninjured, the permeability of the walls is not by that means impeded. We know that the soluble salts of the blood-cells permeate the cell-walls; hence it would be very remarkable if the soluble coagulable protein-bodies of the cell-contents could not also partially penetrate the cell-membranes after the removal of all the serum, in accordance with the laws of endosmosis. Moreover the substance retained in the blood-corpuscles (as C. Schmidt has shown) loses potash by its solution in water and subsequent coagulation, and besides this also organic matter; so that this method, even if all the serum could be removed from the blood-corpuscles, would prove insufficient to determine the solid constituents of the blood-cells.

C. Schmidt<sup>1</sup> is the first who has attempted the solution of the problem, to determine the relation of the moist blood-cells to the intercellular fluid. His mode of proceeding is not based, as might be supposed, on the direct determination of the dry blood-corpuscles by means of sulphate of soda, but, on the contrary, on the original method of Prevost and Dumas. Since the investigations of the most accurate analysts show that the solid constituents of the serum stand in a constant relation to those of the clot, that is to say, since the richness of the clot in solid constituents is proportional to the degree of concentration of the serum, it follows that the number representing the dry blood-corpuscles, calculated according to Prevost and Dumas' method, must also stand in a constant relation to the fresh corpuscles existing in the blood. It thus became necessary to discover the constant factor by which we might calculate the blood-cells (in the morphological sense) from the hypothetical dry blood-corpuscles found by Prevost and Dumas' method. Schmidt has found that this coefficient is equal to 4, so that we have only to mul-

<sup>1</sup> Charakteristik der Cholera. S. 3-19.

tiply the hypothetical dry blood-corpuscles by 4, in order to obtain the number representing the moist blood-cells. Schmidt's experiments show that a number, larger or smaller by 0·3 than 4·0, fails to give the correct relation. It was chiefly by the three following methods that Schmidt was enabled to determine this factor:—

1. He determined, by micrometrical measurement, the diminution which the red blood-cells undergo on drying. If they are dried under circumstances which admit of a uniform evaporation of water in all directions, Schmidt finds that they undergo a constant diminution of volume, amounting to 68 or 69% of that of the fresh cells; hence the latter contain about 68 or 69 parts of water to 32 or 31 of solid substances, or nearly four times as much of solid constituents as is dissolved in the plasma.

2. After Schmidt had convinced himself that the quantities of serum expressed from the clot at different times had the same density and the same composition, he investigated by the microscope the volumetric relation existing between the blood-cells and the intercellular substance (fibrin + serum) in the clot in its most contracted state; and ascertained that in 100 volumes of clot, there are at the most 20 volumes of intercellular substance, or one-fifth of the whole volume; if, therefore, the four-fifths of the volume of the blood-corpuscles in the clot be compared with the volume of the whole blood (clot + serum), we find that the blood must contain at least 40%, by volume, of fresh cells; Schmidt moreover found, in further comparisons of this kind, that, as a general rule, the blood contains a larger volume of blood-cells, and that it may rise to 53 or 54% of the whole volume.

3. The third equation of condition which Schmidt applied to the determination of this coefficient, depends on the comparison of the unequally divided mineral constituents in the clot and serum. It has been already fully demonstrated that potash-salts and phosphates predominate in the blood-cells—a point on which any one may easily convince himself by comparing an accurately made ash-analysis of the clot or of the cruor (if the fibrin has been removed) with that of the corresponding serum. Since, unfortunately, the serum is never entirely free from phosphates and potash-salts, while the blood-corpuscles are never entirely free from alkaline chlorides and soda-salts (in the analyses made by Schmidt, and in accordance with his method), this might appear to be the best check on the coefficient established by Schmidt; but unfortunately, it cannot be used to ascertain, in a special case, whether Schmidt's calculation of the relation of the cells to the intercellular fluid be correct. If there were a substance to be detected in the serum, so peculiar, and so easily separable and determinable quantitatively, by chemical means, as the hæmatin in the blood-corpuscles, then from the analysis of the clot, and from the quantity of this substance peculiar to the serum, it would be very easy to calculate how much serum (which must obviously also have been analyzed) was enclosed in the clot; if then we deduct the other constituents pertaining to the serum (as determined by the analysis of it), from the quantity of similar substances and the fibrin found in the clot, we should at once obtain, by the simplest calculation, the quantity and the composition of the blood-corpuscles contained in 100 or 1000 parts

of blood. If this were the case, the problem would be completely solved, but, unfortunately, neither in the preformed sulphates nor in the organic matters can we find a substance which is entirely excluded from the blood-cells. Hence, in all probability, we must forever rest satisfied with Schmidt's coefficient, as affording the closest approximation; but if other parts of the blood-analysis were equally accurate, this coefficient would always afford highly correct results. Physiology, and especially physiological chemistry, are indebted for the most brilliant results to this ingenious combination of Schmidt's.

Schmidt's method of calculation in analyzing the blood is very easy of comprehension: we have the analyses of the clot and of the serum, and the proportion (calculated from these data) of the constituents of the whole blood: if we multiply by four the number of the dry blood-corpuscles calculated by Prevost and Dumas' method, we obtain the quantity of fresh blood-cells, and hence their ratio to the intercellular fluid. We now deduct, from the analysis of the whole blood, the constituents belonging to the quantity of intercellular fluid, and the remainder represents all the substances belonging only to the blood-corpuscles.

Vierordt,<sup>1</sup> has recently succeeded, by means of very comprehensive and unusually laborious investigations, in sketching a method of a blood-analysis, which promises to supply some of those deficiencies in Schmidt's method, which have been felt by all experimentalists, and may therefore serve in some degree at least as a check upon the latter. If we were able to determine the numerical quantity and the volume of the blood-corpuscles in every kind of blood to be analyzed, we should naturally have no difficulty in obtaining their relations of weight in the blood, and of determining by a simple calculation from the further analysis of the blood the amount of constituents appertaining to the blood-corpuscles. Vierordt was thus led, at the expense of much time and labor, to calculate the number of the blood-corpuscles in the two following ways. In the one method, a small volume of unmixed blood was measured in a capillary tube, and then introduced into what he termed a diluting fluid (a tolerably concentrated solution of albumen or gum), with which it was spread out under the microscope, and the corpuscles were then counted by means of two glass micrometers, which had been graduated expressly for this purpose, and were respectively attached to the eye-piece and the object-glass. By the other method, an accurately measured volume of blood was mixed with an equally accurately measured volume of diluting fluid, and a microscopic volume of this mixture was employed for the counting of the corpuscles. Welker<sup>2</sup> has recently suggested certain modifications in Vierordt's method.

Vierordt does not determine the volume of the blood-corpuscles by direct measurements, but by a simple calculation, which is perhaps scarcely exact enough. This calculation, as well as that of the whole blood-analysis, depends essentially upon the circumstance that comparative analyses are made of the whipped blood, rich and poor in cor-

<sup>1</sup> Arch. f. physiol. Heilk. Bd. 11, S. 26-78, 327-332, 547-558, and 854-884.

<sup>2</sup> Fechner's Centralbl. 1858. No. 12, S. 218-222.

puscles; according to Vierordt, the blood is rendered poor in corpuscles either by the filtration of the whipped blood through paper, or by the addition of a definite quantity of previously analyzed serum. All persons familiar with the principles of mathematics will readily comprehend Vierordt's ingenious method, although they must at the same time perceive that, for these calculations, besides the exact counting of the corpuscles in each analysis, it is necessary to assume one, if not two, probable magnitudes.

If we abstain from entering more fully into this subject, and for the present withhold our judgment on a question of physiological chemistry which promises to be of the highest importance to physiology generally, this does not arise from a want of appreciation of the great merits of Vierordt in this department of chemistry, but simply because we do not regard a manual of this kind as a suitable place for the introduction of investigations of this nature, and because we have been anxious, as far as possible, to base our judgment solely upon our own experiments and upon *post-mortem* examinations. In consequence of the different direction of our own investigations regarding the blood, we are hardly in a position to criticise in their individual details the labors of Vierordt. Such a critical and experimental testing is, however, indispensably necessary before we can form a correct judgment of this method, as a number of considerations force themselves upon our notice, which, although they have in part been explained by Vierordt himself, are still sufficiently numerous to demand an experimental examination. O. Funke,<sup>1</sup> among others, has shown with much clearness the possible causes of error which appertain to this method. For the present, we may regard Schmidt's method of blood-analysis, of which we have considerable experimental knowledge, as the one which, notwithstanding some well-known deficiencies, affords the most certain results.

No one has yet attempted a quantitative determination of the *colorless blood-cells*; it is probable that we shall never arrive at more than an average estimate of them.

We have already spoken (page 318) of the quantitative determination of the *fibrin*, and pointed out that it is not to be relied upon. We will here only add a few words regarding the results of Hinterberger's experiments, which show that we always obtain less fibrin by whipping the blood than by washing the clot. He is of opinion that the evaporation of the water occurring during the coagulation of the blood, may be one of the causes of this difference; considering, however, the comparatively small quantity of fibrin in the blood, the error arising from this cause would be infinitesimally small, and in the best analyses of the blood would be overbalanced by other errors of observation; thus, one of the first rules of analytical chemistry is, that liquid and volatile fluids which are to be submitted to quantitative analysis should never be allowed to stand, if it can be avoided, even to be weighed in open vessels; hence a specimen of blood intended for a quantitative analysis should never be allowed to coagulate in an open vessel, and then perhaps to stand for 24 hours. But in following this analytical rule, direct experiments show us that we obtain less fibrin from the whipped blood than from washing

<sup>1</sup> Schmidt's Jahrb. d. ges. Med. Bd. 74, S. 1-7, and Bd. 78, S. 5-9.

the clot. We have already shown in this volume, that even the fibrin obtained by whipping, since it can be only imperfectly washed, is never pure fibrin; and this is far more the case with fibrin obtained from the clot: while a little blood-pigment always remains in the former, the latter contains colorless corpuscles and the walls as well as the granular contents of the colored cells. The colorless cells and the walls of the colored cells may often occur in such quantities as entirely to falsify the number representing the fibrin: indeed, in the blood of the hepatic veins we have already become acquainted with a case in which scarcely any fibrin occurs, and where it was merely the cell-membranes of the blood-corpuscles that were mistaken for fibrin. The greater number of the fine flakes which in whipped blood penetrate the linen filter, are cell-membranes of this sort, the flakes of fibrin being in fact comparatively few; the pseudofibrin of the blood of the hepatic veins passes almost entirely through the linen filter. Hence, in an analysis, we have the two alternatives, either of losing some of the fibrin or of simultaneously including in the calculation both colorless blood-corpuscles and cell-walls; hence more fibrin will invariably be obtained from coagulated blood, in which these elements are firmly enclosed by the fibrin, than from whipped blood, whose fibrin (if no water has been added) had been separated by a linen filter. Moreover, linen filters are not to be trusted for a quantitative analysis; for they either allow a number of minute flakes of fibrin to pass through them (whether the fibrin be obtained by whipping, or by kneading and pressing the clot), or they become invested with a fine viscid crust of cell-walls, by which even the widest meshes of the linen become clogged up. Hence, in making as accurate an analysis of the blood as possible, a linen filter should be altogether avoided for the quantitative determination of any of the constituents, and we then find that the fibrin obtained from the clot does not exceed that which is obtained by whipping the blood; and further, that the experiments instituted by Marcchal, and more recently repeated by Corne,<sup>1</sup> on the influence of motion on the diminution of the fibrin, and the deductions they have drawn from them, are entirely dependent on errors in their mode of analysis. It is certainly impossible altogether to avoid these errors; for if we carefully collect all the insoluble substances passing through the linen filter, the number which we obtain for the fibrin is too high. The following is, we believe, the best, although it is by no means a perfect method of collecting all the insoluble matter which collectively goes under the name of fibrin: whipped blood must be strongly watered, and the flakes allowed to deposit themselves (this deposition is, however, often very imperfect); the fluid, as far as it has become clear, is to be drawn off, and the turbid residue, with the coarser clots, to be repeatedly shaken with water, and the clear fluid to be removed till water ceases to be at all colored by it; afterwards, if it be possible, a paper filter must be used, through which the fine flakes cannot penetrate, in place of a linen one; previously, however, heating the fluid with an equal volume of spirit. (When the fluid has once become colorless, no more coagulable substance is generally found in the fibrin.) The fibrin may then be tolerably easily collected on the filter, and it

<sup>1</sup> Compt. rend. T. 30, p. 110.



should afterwards be washed with boiling spirit. We cannot even in this way accurately determine the fibrin; but we know what we have to deal with, and that if we always determine the fibrin in excess, it is far less dependent on pure casualties than if we had used linen filters, or had partially watered the blood; in one case any calculation is impossible, in the other we know that there is always an excess of fibrin and a slight loss of blood-corpuscles. Our knowledge of the error leads us to hope that we may subsequently learn to avoid it: accuracy and patience are indeed indispensable in this mode of determining the fibrin. Moreover, in this somewhat circumstantial operation, the blood does not readily undergo putrefaction, in consequence of the frequency with which the water is changed.

For the method of determining the *albumen* in the serum, we must refer to the observations already made in p. 304. In regard to the special application of those remarks to the analysis of the blood, we need only add that it is always important, besides determining the albumen in the serum, to determine also the coagulable matter contained in the clot, or the clot when free from fibrin, or in the defibrinated blood, as a controlling check which is indispensable in blood-analyses; so as in some degree to combine the methods of Becquerel and Rodier, or of Popp, with that of Scherer. Hinterberger found that the amount of albumen, when determined according to Becquerel and Rodier (by extraction of the solid residue of the serum with various indifferent solvents), was always somewhat larger than when determined by Scherer's method; this is, however, not merely the case with the serum, but also, in a still higher degree, with the cruor when free from fibrin (the blood-corpuscles + the enclosed serum); that is to say, here also the substance obtained by coagulation through the aid of acids, amounts to less than the residue which we obtain after treating the solid constituents of the cruor with ether, alcohol, and water. This result, which can be observed in any analysis of the blood, partly depends upon the circumstance that when the coagulation is effected by the aid of acids, they extract from the coagulable matter a small quantity of earths which would naturally remain in the substance, if treated only with indifferent menstrua; but partly also on this, that, by the treatment with such menstrua certain alkaline salts, and probably also organic matters, are extracted from the residue, which, in the coagulation, retain in solution a certain quantity of albuminous substances from the fluid, so that this portion is abstracted from the coagulation.

A. Becquerel<sup>1</sup> has recently availed himself of Biot's discovery of the rotatory power which dissolved albumen exerts on polarized light, in order to determine the quantity of albumen dissolved in the serum, as Bouchardat had previously attempted to do. We cannot give a minute description of the instrument in this place, but we may observe that it enables us to measure the rotation which a pencil of light undergoes to the left hand in consequence of the albumen contained in the fluid. According to Biot's formula, the rotatory power of albumen is  $27^{\circ} 86'$ ; in Becquerel's apparatus, the *albuminimeter*, each minute of deviation which the pencil of light undergoes, corresponds to 0.18 of a gramme

<sup>1</sup> Compt. rend. T. 29, p. 625.

of albumen in the solution enclosed in the apparatus, and hence every degree corresponds to 10·8 grammes. Becquerel has found, by repeated observations, that there is a perfect coincidence between this physical and the chemical analysis; but this is a subject requiring further investigation; in the first place, because, according to Becquerel's admission, even the chemical mode of determining the albumen does not give strictly accurate results: and secondly, because the serum always contains traces of sugar, which may, to a certain degree, modify the amount of the deviation.

The determination of the *salts* of the serum and of the *crur* is best accomplished by carbonizing the solid residue of each, and then adopting the modification of Rose's method, which is described in p. 369. The salts can then be analyzed in accordance with the rules recently laid down by Rose, whose labors have gone far to bring this department of analytical chemistry to a state of perfection.

The quantitative determination of the *fats* in the blood, as in other animal substances, is associated with difficulties which often cannot be entirely overcome. As a matter of course, we only employ for this purpose the solid residue, after thorough drying at 120°. The best method of procedure is to introduce into a small digesting flask the dry substance which is to be employed for the determination of the fat, while we determine its weight, as in elementary analyses, by re-weighing. A small digesting flask is necessary for this purpose, since it is only thus that we can boil the substance with ether, and pour off the ethereal fatty solution without loss of fat. The ethereal solution is then to be evaporated from a small glass cup or basin with a very high border, because the fat very readily creeps up to the edges, and thus necessarily occasions loss. Moreover, the ether must be perfectly pure,—as free as possible from water, alcohol, and free acid. The evaporation of the ether must be accomplished without boiling; and the fatty residue, like all other residues, be dried at 120°.

Pure ether extracts only the neutral fats and the free fatty acids, and not the *alkaline salts of the fatty acids*; the latter must be extracted with absolute alcohol, to which about one-tenth of its volume of ether has been added. The determination of the soaps is always uncertain, because, as a general rule, we do not obtain them in sufficiently large quantities to separate the non-fatty substances which almost invariably intermingle with them.

To calculate the *extractive matters* by deducting from 100 parts of the fluid the sum of the constituents obtained by direct analysis, is a procedure by no means to be recommended; for by such a course we lose one of the most important means of checking or controlling the whole analysis. After the removal of the fats from the solid residue which we are going to analyze, the extractive matters, that is to say, the alcoholic, spirituous, and watery extracts, must be dried, weighed, and finally incinerated, in order that the ashes (after their determination) may be extracted from the organic matter; it is only in this manner that we can hope to attach any scientific value to these extractive matters, which, in a physiological point of view, are doubtless of much importance.

We certainly cannot apply all the controlling checks which we have here described in every analysis of the blood; but as the little regarded rule holds good (see p. 408), that the smallest possible quantities give the most accurate results for each individual determination, by no means so large a quantity of material is requisite for a good analysis of the blood, as we are commonly in the habit of supposing necessary. It is only for analyses of the ash, that larger quantities are required, and even here the accuracy attainable in inorganic analysis is now so great, that a comparatively small quantity suffices.

*Sugar and urea* may sometimes be determined quantitatively in the blood; on these points it is sufficient to refer to what has been already stated in p. 484, and pp. 148 and 152.

In relation to the *determination of the specific gravity*, we shall more fully notice this subject in the chapter on "the urine," when we shall examine the different methods which have been proposed for this purpose. With regard to the blood, we need only remark that it is very often almost impossible to determine the density of the cruor free from fibrin, and of the defibrinated blood, in consequence of the viscidness of this fluid, and of the air-bubbles suspended in the blood, and, in particular, adhering to the vessel.

A full consideration of all the circumstances and accidents appertaining to chemical analysis, must shake our confidence in the relative accuracy of those analyses of the blood of which we are at present in possession, and we might even hesitate in ascribing any degree of value to the deductions and hypotheses which have incautiously been drawn from them. Then, moreover, it must be remembered that in many diseases in which the admixture of the blood is most altered, good analyses of blood cannot, from considerations of humanity, be adequately prosecuted, and that in reporting such analyses, we have generally been contented with a vague and abstract diagnosis, although the course of the individual morbid process is of the highest importance in a scientific point of view: hence no great weight can be attached to a humoral pathology which is based on such slender supports. If, finally, we consider that in all kinds of analyzed blood, the result refers only to the greater or lesser fluctuations in the relations of the main constituents of the blood, and not to a new alteration, admixture, or decomposition of that fluid, and since these relations have not yet been adequately elucidated in a chemical point of view, we can only wonder that it should ever have been supposed that such scanty materials could aid us in obtaining any insight into the obscure mysteries of morbid processes. We will not deny that thanks are due to those who have prosecuted the most comprehensive investigations with minute carefulness and disinterested labor; but we should be untrue to the cause of science, did we fail to set forth the real character of these results.

We have already attempted, at the beginning of this chapter (see p. 546), to give a general view of the *quantitative composition of the blood*; and we will now proceed to consider the varying proportions of the individual constituents under different physiological and pathological conditions.

The ratio of the *blood-cells* (in the morphological sense) to the inter-cellular fluid, appears to undergo very slight fluctuations in the normal

state when the physiological conditions are analogous. In an adult healthy *man*, we find on an average 512 parts of moist blood-corpuscles in 1000 parts of blood; the fluctuations do not exceed a difference of more than 40 in either extreme, so that while 472 would be a very low number, 552 would be a very high one for the proportion of cells in the blood of a man.

Vierordt<sup>1</sup> found in his various countings that in  $\frac{1}{1000}$  cubic millimetre [the linear millimetre being about  $\frac{1}{25}$ th of an inch] of normal blood obtained by pricking the finger, there are on an average 5,055,000 blood-corpuscles; Welker,<sup>2</sup> on the other hand, fixes the number at 4,600,000.

According to the above-described method, the dry blood-corpuscles found by Prevost and Dumas, amounted to 129 p.m., by Lecanu<sup>3</sup> to 132.5, by Andral and Gavarret<sup>4</sup> to 127, by Richardson<sup>5</sup> to 134.8, by Becquerel and Rodier<sup>6</sup> to 141.1, by Nasse<sup>7</sup> to 116.5, by Popp<sup>8</sup> to 120, and by Scherer<sup>9</sup> to only 112.

It is scarcely necessary to observe that no conclusion regarding the proportion of the cells of the plasma can be drawn from the proportion of the serum to the clot: as we have already seen in the preceding remarks the sinking capacity of the blood-corpuscles on the one hand, and the contractility of the fibrin on the other, present such variations that we may readily comprehend how one voluminous clot may contain very few blood-corpuscles, whilst another which is less voluminous may contain a proportionally larger number of cells.

In the blood of *women* we find on an average fewer cells than in *that* of men; their number is still more decreased during pregnancy, before the period of menstruation, and after its entire cessation towards the close of the climacteric period.

We are especially indebted for the determination of these relations to Becquerel and Rodier, who give 127.2 as the mean number for the corpuscles of the blood of women. Nasse, in his experiments on the blood of animals, has found the same differences in the different sexes.

Prevost and Dumas,<sup>10</sup> Berthold<sup>11</sup> and Simon<sup>12</sup> have shown by direct investigations, as might indeed be conjectured, that the number of blood-corpuscles varies in the *blood of different animals*; and more recently the same subject has been fully considered, especially by Nasse,<sup>13</sup> but likewise by Andral, Gavarret, and Delafond.<sup>14</sup> According to these researches, it would appear that the cold-blooded animals contain far fewer blood-cells than those having warm blood, birds on an average more than mammalia, but carnivorous not more than herbivorous animals. The blood of the pig contained relatively the largest number of cells.

<sup>1</sup> Arch. f. physiol. Heilk. Bd. 11, S. 867-874.

<sup>2</sup> Fechner's Centrabl. 1853. No. 12, S. 222.

<sup>3</sup> Etudes chimiques sur le sang humain. Paris, 1837.

<sup>4</sup> Recherches sur les modifications de quelques principes de sang, &c. Paris, 1842.

<sup>5</sup> Thomson's Record of General Science, vol. iv. pp. 116-135.

<sup>6</sup> Recherches sur la composition du sang, &c. Paris, 1844.

<sup>7</sup> Op. cit. <sup>8</sup> Op. cit. <sup>9</sup> Op. cit.

<sup>10</sup> Bibliothèque nouvelle, T. 4, p. 125.

<sup>11</sup> Beiträge zur Zoologie, u. s. w. Göttingen, 1831.

<sup>12</sup> Lehrb. d. Ch. Bd. 2 S. 285 [or vol. i. p. 839 of the English translation].

<sup>13</sup> Handwörterbuch der Physiologie. Bd. 1, S. 188; and Journ. f. prakt. Ch. Bd. 28, S. 146.

<sup>14</sup> Ann. de Chim. et de Ph. 3me Sér. T. 5, p. 304.

Nasse found in the blood of the pig 145·5 p.m. of dry blood-corpuscles, in that of the hen 144·6, of the goose 121·4, of the dog 123·8, of the ox 121·8, of the horse 117·1, of the cat 113·4, of the calf 102·5, of the sheep 92·4, and in that of the goat only 86·0 p.m. The results obtained by the other inquirers can only be compared with one another, but do not admit of a comparison with those of others. It is worthy of notice that Prevost and Dumas found the corpuscles in the blood of the land-tortoise to be very abundant, and even relatively more numerous than in the blood of the duck, the raven, and some of the mammalia. The correctness of these numbers ought to be investigated, since land-tortoises bear great affinity in an anatomical point of view to birds, whilst sea-tortoises stand in a nearer relation to fishes.

It may be shown with tolerable accuracy, that the quantity of the corpuscles is not the same in *the blood of all the vessels*; for when, for instance, the urinary secretion is very active, the venous blood in the kidneys will contain relatively more corpuscles than the arterial blood of those organs. In consequence of the essential differences which take place in the blood-corpuscles of the spleen, the venous blood of this organ is found to differ from the arterial, not only qualitatively, but also quantitatively, in reference to the blood-cells. We learn from the investigations of Mayen, Hering, and Nasse, that the arterial blood contains fewer blood-corpuscles than the venous. Schmid found a much smaller number in the portal blood than in that of the jugular veins; I found a much larger quantity in the blood of the hepatic veins than in that of the portal vein, and even more than in that of the jugular veins, the vena cava, and the splenic vein.

In the blood of the hepatic veins of a horse which had been fed four hours before death, I found 743 p.m. of moist blood-cells, whilst there were only 592 in the blood of the external jugular vein of the same animal, only 664 in that of the vena cava, 573 in that of the portal vein, and only 322 in that of the splenic vein.

My own experiments, as well as analogous physiological observations, concur in showing that *scanty nutrition* and *prolonged abstinence from all food* diminish the number of the blood-corpuscles.

From what has been already said in reference to the function of the liver (see page 491), and the influence of fat on the formation of cells (page 236), we need not wonder that Popp should have found an augmentation of the number of the blood-corpuscles, and more especially of the colorless ones, *after the prolonged use of cod-liver oil*.

We should naturally expect that repeated venesections would occasion a diminution in the number of blood-corpuscles; and Andral and Gavarret, Simon, Becquerel, and Rodier, Zimmermann, Popp, and Nasse, have shown by direct experiments that this is the case. Although the correctness of these views has been proved by all the inquiries instituted on the subject, no average proportion has as yet been established between the diminution of the blood-corpuscles and the quantity of the blood abstracted, or the number of times venesection had been performed.

We cannot hope to discover a definite proportion between the decrease of the blood-cells and the abstraction of blood, until we can accurately determine the individual magnitudes of all the coincident momenta. It

is not difficult to perceive, that for the present, no such determination can be arrived at; for the intercellular fluid will in one case (as for instance, from deficient nutrition in already depressed and reduced organisms) be less rapidly regenerated than in another, and on this account there will be a less marked difference between the blood-cells and the plasma; whilst, on the other hand, the blood-cells may, under favorable conditions, be more rapidly reproduced, and in that case also, the relation between the plasma and the cells would be less unequal. Finally, it may happen that the blood-cells are more rapidly destroyed in one organism than in another, and hence the difficulty of determining these physiologically important relations is increased. Moreover, experiments of this kind have usually been instituted during the manifestation of morbid processes whose various characters and modes of development have not been taken into consideration.

Since the colored blood-cells, as we shall subsequently show, are produced from colorless cells, it is not surprising that after repeated or very copious venesections (Remak) the ratio of the colorless cells to the colored ones should be considerably increased, or, at all events, that the former should be less diminished in number than the latter.

It has even been found, that during different periods of one and the same bloodletting, the relation between the blood-cells and the plasma is not always constant. Becquerel and Rodier, who specially investigated this subject, did not arrive at any definite numerical relations. In the great majority of cases, the corpuscles were diminished in the blood, which flowed last, but sometimes they were increased. No light, however, can be thrown on this subject as long as we remain in ignorance of the physical relations existing between the relative amounts of the blood and of the other juices of the animal body.

It will be long before we can hope to establish any fixed relations of comparison between definite physiological processes and the increase or diminution of the number of blood-corpuscles in morbid blood. We constantly find the blood-cells augmented in plethora, in the earlier stages of heart-disease, in spinal irritation (Popp), and in cholera (C. Schmidt). It may be readily conceived that a diminution in their numbers is of more frequent occurrence, especially in those anæmic conditions which generally supervene upon profuse diarrhoeas, prolonged suppurations, slow intermittent fever, typhus, copious exudations, exuberant morbid growths, cerebral affections, chronic metallic poisonings, and other severe diseases; in short, in all cases where the formation of the blood is less than its consumption. In chlorosis, which properly speaking is only an anæmic condition, and which, from our ignorance of its immediate cause, has been termed spontaneous anæmia, the colored blood-cells are extraordinarily diminished, although Becquerel and Rodier state that they have observed two cases of this disease in which the chlorotic blood was rich in corpuscles. During the first eight or ten days of typhus, the blood-corpuscles are always increased; but subsequently to that period, at least until the twenty-first day, their number is considerably diminished. In other diseases, we are unable to trace any very perceptible fluctuations in the number of the corpuscles; and hence the results of most experimentalists do not coincide very closely.

Becquerel and Rodier, as well as Popp, agree however in asserting that the number of blood-corpuscles is diminished in violent inflammations, pneumonia, and acute articular rheumatism.

In *chlorosis*, the amount of the dry blood-corpuscles has been found to sink to 80, and even as low as to 46·2 p.m. In *spinal irritation*, Popp found 120·5 p.m. as the lowest number, and 140·5 p.m. as the maximum (his mean normal number being 120); in *plethora* he found the corpuscles much less increased than in spinal irritation. Schmidt, who found 513 moist blood-cells in normal male blood, saw the number rise to 559 in cholera; in the blood of women (where the mean number for the corpuscles is about 400 p.m. according to him), the number has risen to 464. The bare results of the analyses cannot attain any physiological value until we are able to determine the conditions in which this augmentation of the corpuscles is absolute, or in which it is merely relative; and for the present we can only hazard a conjecture in reference to this question. In cholera the apparent augmentation is only relative; for the admirable investigations of Schmidt and others on the blood in cholera show that in this disease water and salts are the principal constituents which are lost, that the serum is consequently thickened, but diminished in volume, and that its ratio to the blood-cells is therefore also diminished. Moreover, according to Schmidt's calculation, a number of corpuscles are destroyed in cholera, so that the blood of a healthy individual contains absolutely more blood-cells than that of a cholera patient.

" In the earliest stage (within the first week) of typhus, as well as in plethora and spinal irritation, we are inclined to believe that there is an absolute increase of cells; at all events, no separation of serum, or of any of its constituents, is ever observed in any of these conditions.

We forbear entering any further into the detail of the observations made on the proportion of the corpuscles to the intercellular fluid in diseases, since they have not led to such complete and available results as to justify us in more fully noticing this subject without some deeper insight into the individual pathological processes.

Very little is known in regard to the alterations which the *chemical composition of the blood-corpuscles* undergoes under different physiological and pathological relations, since no light has hitherto been thrown upon their morphological condition; inquirers having limited themselves to an attempt to determine the frequently mentioned dry blood-corpuscles, without reference to the water appertaining to them, or to the soluble constituents which they may contain. If we were to attempt to calculate these relations from the older analyses, we should readily be led into error, even if our calculations were not wholly impracticable; since all the relations cannot be taken into account when we draw deductions from an investigation which has been entered upon from wholly different points of view. We regard a mere re-calculation of the analyses as of little or no use, unless they are tested by others conducted by better methods, and prosecuted from a different point of view. Science presents therefore, very few materials for the comprehension of the modifications exhibited in the composition of the blood-corpuscles.

The *amount of water in the blood-cells* undoubtedly stands in a definite relation to the amount of water in the serum, as we may easily

perceive from the morphological behavior of the blood-cells on the addition of water, or dilute or concentrated saline solutions. In this point of view, the blood-cells, moreover, are constantly reacting on the intercellular fluid. Then, further, it is easy to perceive that the constituents of the blood-cells, which differ essentially from those of the plasma, must also have different degrees of diffusibility for water; and that the quantity of water contained in the blood-cells must always differ from that in the intercellular fluid. We have already seen, from Schmidt's investigations, that the solid constituents of the blood-cells are almost four times as great as those of the serum; that is to say, if 100 parts of blood-cells contain 32 solid parts, we shall find in 100 parts of serum little more than 8 parts of solid matter. In the meanwhile, the most accurate of Schmidt's investigations regarding human blood, and of my own, on the blood of the horse, by no means present a constant relation between the quantity of water contained in the blood-cells and that in the serum; but such a result was not to be expected from the differences which the cells present in their chemical constitution. This much only is certain, that when the quantity of the water is decreased in the serum, it is likewise similarly decreased in the blood-cells; and in the same manner, when an augmentation occurs in the former, it is also perceived in the latter. From the observations hitherto instituted in reference to morbid blood, it has been believed that we might establish the following general proposition:—that the quantity of water contained in the blood bears an inverse relation to the number of the blood-corpuscles; but the above remarks must have sufficiently shown that this cannot be received without a certain limitation, more especially as the rule presents numerous exceptions. The decrease in solid constituents is not limited in these cases to the solid substances of the blood-cells, but extends in a corresponding proportion to those of the serum. It is evident that where there is an absolute diminution of the blood-cells and an increase of the serum, the blood must, on the whole, be richer in water when the heavier morphological elements are diminished. When we treat of the serum, we will enter more fully into the relations on which the greater or lesser quantity of water in the blood depends.

The composition of the blood-corpuscles differs, moreover, in respect to their proximate solid constituents. We have seen that *globulin* and *hæmatin* do not stand in a definite numerical relation to each other in the colored blood-cells. The hæmatin of different animals appears, from Mulder's researches, to be perfectly identical; and we might, therefore, draw a conclusion from the iron contained in the blood-corpuscles regarding the quantity of hæmatin. From Schmidt's calculations, which have been partly based upon direct investigations, and partly on the analyses of others, it would appear that, for every one part of metallic iron, there occur in the blood of men 230 parts of corpuscles (according to Becquerel and Rodier, 251); in the blood of women, 229; in that of oxen, 196.5; in that of pigs, 223; and in that of hens, 307. In the first stage of typhus, where the number of blood-corpuscles is increased, Schmidt found the proportion as 1 : 220, and hence the quantity of the hæmatin was diminished. In those conditions, however, in which the number of the blood-corpuscles is diminished, the hæmatin is relatively



increased; for he found that on an average the relation between the iron and the dry blood-cells was, in pneumonia, as 1 : 248; in chlorosis, as 1 : 269; and in pregnancy, as 1 : 249. In the same manner, Schmidt made the important observation already referred to, that the blood-corpuscles become poorer in globulin and richer in hæmatin after repeated venesections, when the blood has become more watery.

Schmidt has drawn up the following table from cases in which three venesections were performed. The first was a case of pneumonia, in which the blood was analyzed by himself; the second was one of tuberculosis; and the nature of the third is not specified. The analysis in the last two cases was made by Becquerel and Rodier.

	Pneumonia.	Tuberculosis.	Unspecified case.
1st venesection, . . . .	248 : 1 . . .	256 : 1 . . .	252 : 1
2d, " . . . .	233 : 1 . . .	252 : 1 . . .	247 : 1
3d, " . . . .	221 : 1 . . .	234 : 1 . . .	212 : 1

The relative quantity of iron contained in the blood-corpuscles increases, therefore, with each successive venesection. This phenomenon admits of a simple solution, for it would appear, from all observations, that the hæmatin cannot permeate the walls of the blood-cells, which, however, admit of the permeation of their albuminous contents: now if the blood loses solid constituents, the serum becomes richer in water; a diffusion-current of a more diluted solution then enters the blood-cells, whilst a more concentrated stream passes outwards from them; now since hæmatin cannot penetrate through the cell-wall, the loss of solid constituents from the blood-cell must mainly affect the globulin, whilst the hæmatin will, under such conditions, appear to be relatively increased in relation to the globulin + the cell-membrane.

I found the quantity of hæmatin in the cells of the arterial blood of the horse somewhat more considerable than that contained in the blood of the external jugular veins; whilst, on the other hand, the quantity of hæmatin contained in the blood-cells of the hepatic veins is far smaller than that of the portal blood.

I found the ratio of iron to the dried blood-corpuscles in the arterial blood of the horse, as 1 : 394; in the jugular vein, as 1 : 390; in the portal vein, as 1 : 312; and in the hepatic veins, as 1 : 500; these being the mean results obtained from several experiments. The smaller quantity of hæmatin contained in the cells of the arterial blood, compared with those of the jugular venous blood may be referred not only to the greater richness in fat of the arterial blood, but more decidedly, or even exclusively, to the loss of fat which takes place during the arterialization of venous blood by the process of respiration.

In speaking of the formation of blood-cells in the liver, we drew attention to the fact that a small portion of the iron which was conveyed to that organ with the blood-cells of the portal vein, is separated with the bile, while the remaining portion appears to be equally distributed among the blood-corpuscles which have been newly formed within the liver, so that the iron of 100 blood-corpuscles of the portal vein is distributed over nearly 150 corpuscles of the hepatic veins: consequently the blood-cells of the hepatic veins must contain one-third less iron than that of the portal vein.

The blood-corpuscles must necessarily also exhibit differences in their amount of *fat*, since the quantity of fat contained in the blood of different animals and of men, under different conditions, is extremely variable. In reference to this question, I have directed special attention to the differences in the quantity of fat contained in the blood-corpuscles of different vessels of the same animal; and the results of my investigations, taking the mean of several experiments, are, that 100 parts of *moist* blood-cells from the carotid artery of a horse contain 0.608 of fat; 100 parts from the external jugular vein, 0.652; from the portal vein, 0.752; and from the hepatic veins, 0.684. These experiments warrant us in hoping that a further prosecution of such inquiries may throw a very considerable amount of light on the metamorphosis of fat, and on the function of the blood-cells. The first step seems, at all events, to have been made towards the elucidation of that chemical metamorphosis which the blood-cells experience in the pulmonary capillaries by the action of the inspired oxygen.

That the blood-corpuscles contain variable quantities of *soluble salts*, is a fact that is made evident by the above-mentioned investigations of C. Schmidt, in which he ascertained the different proportions of the potash salts and phosphates in the blood-cells to the soda and chlorine compounds in the serum of the blood of different species of animals. But I also found that the quantity of salts contained in the cells of the blood from different vessels of the same animal constantly differed; thus, for instance, 100 grammes of fresh blood-cells from the temporal artery of a horse contained 0.806 of a gramme of salts (independently of the peroxide of iron in the ash); the same quantity, taken from the external jugular vein, contained 0.632, from the portal vein 0.729, and from the hepatic veins 0.893. There is therefore a very considerable difference, in reference to their amount of salts, between the cells of the arterial and those of the venous blood; the former containing more salts than those of ordinary venous blood. The relation between the cells of the portal vein and those of the hepatic veins is still more striking; for although the serum of the portal blood is far richer in salts than that of hepatic venous blood, the difference in the amount of salts in the cells of the two kinds of blood is still more strongly marked.

This excess of saline contents in the arterial blood-cells can only be explained by the loss of other substances, as, for instance, fat and perhaps also extractive substances, which are lost by the venous cells in their passage through the capillaries of the lungs; this increase of the salts during the arterialization of the blood-cells is, therefore, probably only a relative one. The case is wholly different with respect to the saline contents of the cells of the blood flowing to and from the liver. If, as would seem probable from our investigations on the subject, new corpuscles are actually formed in the liver, it follows from this fact that the younger blood-corpuscles contain more salts and less hæmatin than the older cells of the blood of other vessels, and that a certain quantity of salts passes from the serum of the portal vein into the blood-cells of the hepatic veins. This increase of salts in the cells of the blood of the hepatic veins is principally limited to phosphates and chlorides, as I can constantly found in three comparative investigations. In 100 parts of

fresh blood-cells of the portal blood I found, on an average, 0.1593 of chlorine and 0.0578 of phosphoric acid in combination with alkalis; while 100 parts of cells from the hepatic veins contained 0.1796 of the former and 0.0611 of the latter.

Schmidt's investigations on the constitution of the blood during *excessive transudative processes* have thrown considerable light on this subject, and shown the differences manifested in the quantity of salts contained in the blood-cells. In cholera, where the blood loses large quantities of salts in addition to water, the blood-corpuscles are also implicated. The intercellular fluid especially loses large quantities of water and chloride of sodium: and this fluid reacting on the blood-cells, abstracts not only a portion of their water, but also a portion of their salts. As the potash compounds and the phosphates predominate in the blood-cells, it is these salts which chiefly escape into the plasma; consequently these compounds are more abundant in the serum, in cholera than in a state of health. Hence in cholera, the blood-corpuscles become relatively richer in solid organic matters, while they lose a portion of their soluble salts. Schmidt found that in the blood-cells of healthy blood the ratio of water to the solid constituents was as 2.14 : 1; in the blood of cholera patients as 1.77 : 1; that the ratio of the organic to the inorganic constituents in the cells of healthy blood was as 40 : 1, and in the blood in cholera as 58 : 1. Schmidt, moreover, found an analogous and only gradually differing relation in the blood after the administration of drastic purgatives, since here the mechanical metamorphosis corresponded entirely to that set up by the cholera process. In other transudative processes, where the loss experienced by the blood principally affects the albuminates and consequently the organic matters (as in dysentery, Bright's disease, and dropsy from different causes), Schmidt found precisely opposite relations; for the blood-cells present this analogy with the plasma, that while the organic matters decrease in quantity, the relation of the mineral substances to the water remains nearly the same. The ratio of the water to the solid constituents in the blood-cells may be as high as 2.4 : 1, while that of the organic to the inorganic substances may be as high as 28 : 1. The salts, however, according to Schmidt's investigations, remain in the same relative proportions to one another in the cells of blood of this kind as in those of healthy blood.

Very little is known positively regarding what are called the *extractive matters* of the blood-cells; in 100 parts of fresh cells of horses' portal blood, I found on an average 0.482, and in those of hepatic venous blood 0.988 of extractive matters free from salts. We shall find that these substances occur in a far larger quantity in the intercellular fluid of the hepatic venous blood than in that of the portal blood.

[The proportion of colorless to the colored corpuscles in healthy blood has been variously stated. R. Wagner gives the ratio as 1 : 8 or 1 : 10 (now proved to be incorrect); Henle makes the ratio as 1 : 80; Donders and Moleschott subsequently showed that 1 : 373 was about the average ratio. Moleschott<sup>1</sup> has recently experimented upon seven individuals of different ages, with the following results. The numbers are in every case the mean of several countings.

<sup>1</sup> Wien. Med. Wochenschr. No. 8. 1854.

In children from 2½ to 12 years, . . . . .	1 : 226
In young men from 21 to 22 years, . . . . .	1 : 330
In men, " 30 to 50 " . . . . .	1 : 846
In old persons " 60 to 80 " . . . . .	1 : 381
In young women from 14 to 38 years, when not menstruating, . . . . .	1 : 389
In young women when menstruating, . . . . .	1 : 247
In " " when pregnant, . . . . .	1 : 281

—G. E. D.]

As the red corpuscles are rendered invisible by the addition of water to the blood, we may in this manner form an approximate estimate of the quantity of the colorless corpuscles; in the inflammatory crust, however, this may be best done by acetic acid, which renders the fibrinous coagulum perfectly transparent under the microscope, and makes the cells embedded within it much more distinctly apparent. Their quantity in the blood is considerably increased during digestion; after fasting, they almost entirely disappear; at all events this may be observed in frogs kept without food. A diminution in their number is less rarely observed than an augmentation. [The view formerly held by Donders and Moleschott, that the colorless corpuscles increase shortly after taking food, and diminish on fasting, is confirmed by more recent observations independently made by Moleschott, who especially finds that food rich in albumen increases their number much more considerably than food poor in that substance.—G. E. D.] Remak noticed their extraordinary increase after copious venesection. According to Nasse and Popp, their number is often considerably increased in pneumonia and tuberculosis, but this is not constant; in typhus and chlorosis they do not appear to be sensibly altered in quantity. In pyæmia, these cells are certainly often very considerably increased in the blood, but this augmentation has been rather inferred from the so-called metastatic abscesses, than directly observed. The blood is, however, sometimes found to contain a great number of colorless corpuscles in conditions in which there cannot be any purulent resorption, as, for instance, in the case of dogs affected with cutaneous eruptions.

It is principally in the disease first recognized by Virchow, and named *leucæmia* by him [and independently discovered by Bennett, who terms it *leucocythæmia*], that we find a very great augmentation of the colorless corpuscles, their ratio to the colored ones being often as 1 : 3; they consequently communicate a pale-red color to the blood.

Moreover, the blood of the splenic vein is richer in colorless cells of various forms than that of any other vessel, as has been especially shown by Funke.<sup>1</sup> It has been already mentioned that I found the blood of the hepatic veins much richer in colorless cells than that of the portal vein.

We will now proceed to the consideration of those modifications in the chemical composition of the intercellular fluid, which have been observed to occur under different physiological and pathological relations, and will endeavor to arrange them according to the augmentation or diminution of the individual constituents. We will begin with the *fibrin*; but as we have already considered the most important points

<sup>1</sup> Diss. inaug. Lips. 1850; and Zeitschr. f. rat. Med. N. F. Bd. 1, S. 172–218.

in reference to this subject, little more remains to be noticed regarding it.

Opinions are not wholly agreed as to the quantitative difference of the fibrin in *venous* and in *arterial blood*, although Lecanu and Nasse coincide in believing that arterial is richer in fibrin than venous blood; and in this respect their opinion corresponds with my own experiments on horses' blood, in which I found 6·814 p.m. of fibrin in the arterial blood, and 5·384 p.m. in the jugular venous blood; but while the quantity of fat in the blood-cells and in the serum differed in both kinds of blood, it was almost perfectly equal in the arterial and in the venous fibrin (namely, 2·154% in venous and 2·168% in arterial dry fibrin). I found rather more ash (2·172%) in the fibrin of arterial than in that of venous blood (1·907%). The fibrin of arterial blood coagulates more rapidly than that of venous blood. In both kinds of blood, when taken from the horse, there is generally formed a superficial layer of fibrin; but this is much more extensive in venous blood, and also more distinctly limited by the clot, than in arterial blood, which may perhaps be mainly owing to the more rapid coagulation of the arterial fibrin, rather than to the less rapid sinking of the cells of the arterial blood, for these are specifically heavier than the venous blood-corpuscles (being poorer in fat and richer in hæmatin), and should therefore sink more rapidly.

In reference to the difference in the quantity of the fibrin contained in *portal* and in *hepatic venous blood*, I may simply remark that, as has been already stated, I found from 4 to 6% of fibrin in the portal blood, whilst in hepatic venous blood there were only traces of fibrin, and sometimes no *true* fibrin whatever. In the fibrin of the portal blood I found from 6·1 to 7·8%, and F. C. Schmid from 7·4 to 8·7% of fat.

Schmid describes the fibrin of the portal blood as a greasy, viscid, or gelatinous mass. In horses which had been fed some (5 to 10) hours before death, I found the fibrin precisely similar in character to that of jugular venous blood; it likewise always formed a very dense and consistent crust in coagulated portal venous blood. Moreover, I could not discover that this fibrin was very readily soluble in a solution of nitre. We have already spoken, in p. 319, of the different amount of fibrin contained in the blood in diseases. It appears, from the most recent analyses of Becquerel and Rodier,<sup>1</sup> that the amount of fibrin may vary very considerably in the same group of diseases, in one case rising above and in another falling below the mean number; as, for instance, in dropsies, and in the most various forms of heart-disease; in chlorosis the quantity of fibrin is either normal, or amounts to 0·1 or 0·2% above the normal quantity; the reverse is the case in anæmic conditions, in which we frequently observe a diminution of this constituent. While no increase of the fibrin was observable in the acute form of Bright's disease, chronic cases of that disease presented an almost constant augmentation of this constituent.

In scurvy Becquerel and Rodier found a constant diminution of the fibrin, but unfortunately these writers have designated as scorbutic that condition in which, in consequence of other grave diseases, the fibrin of the blood falls below 0·2%; on the other hand, in acute idiopathic

<sup>1</sup> Gaz. Méd. de Paris. 1852. No. 24, 25, 26, 30, 31.

scurvy, there was rather an augmentation of the fibrin. As long as such a want of clearness appertains to our ideas of certain diseases, and their various characteristics are so unsystematically confounded, pathological chemistry can make no positive advance, notwithstanding all the efforts devoted to the study of this branch of science. It seems to us that Becquerel and Rodier would have done far more to advance pathology if they had investigated the excretions and some of the secretions conjointly with their analysis of the blood in any single patient, instead of making numerous and laborious determinations of the blood in similarly named but not analogous morbid conditions.

We will now proceed to consider the *constituents of the serum*, and in the first place the *quantity of water* which it contains in different conditions. On this subject we are also indebted to Nasse for our most accurate information. We need not here again repeat that the quantity of water in the serum influences the quantity in the blood-cells, and that consequently the following statements, regarding the augmentation or diminution of the water, may be regarded as referring to the whole mass of the blood. All experimenters without exception, concur in the statement that the serum of *women* is richer in water than that of *men*; and the most recent comparison of the two kinds of blood (that, namely, by Schmidt) yields the same result; in the serum of man's blood Schmidt found 90·884%, and in that of woman's blood 91·715% of water. In *pregnancy* the blood is still richer in water. Serum obtained from the *placenta* contains, according to Poggiale,<sup>1</sup> less water than that from *new-born infants*; the blood of new-born infants, however, contains less than that of adults; in old age the quantity of water again visibly rises. Nasse, on the other hand, found that the blood of the embryonic animal was richer in water than that of the mother.

In different *animals* the quantity of water in the serum and in the blood presents considerable variations; Prevost and Dumas, Berthold, Nasse, and more recently Poggiale, have instituted extensive series of comparative investigations; notwithstanding many differences in individual details, the results of these observers coincide in the following points: namely, that the serum of the *amphibia* contains the largest amount of water, and that of *birds*, on an average, a larger quantity than that of the *mammalia*; and that of the latter class, the serum of *swine* contains the least, and that of *goats* and *sheep* the most water.

In regard to the quantity of water in the serum of blood from *different vessels*, the following may at all events be laid down as a general rule: the serum of *arterial blood* is more watery, and hence specifically lighter, than that of *venous blood*, according to the experience of most observers (although Lecanu and Letellier maintain the contrary); in the serum of the blood of the temporal artery of a horse I recently found 89·333%, and in that of the external jugular vein 86·822% of water. Zimmermann<sup>2</sup> found the serum of the veins of the lower or hinder extremities (of men and animals) poorer in water than the upper or anterior ones.

The *serum of the portal vein* is richer in water than that of any other vein; according to the unanimous opinions of Schultz, Simon, and

<sup>1</sup> Compt. rend. T. 25, p. 198-201.

<sup>2</sup> Arch. f. phys. Heilk. Bd. 6, S. 587-600.

F. C. Schmid, who have all experimented on the subject. My own experiments lead me to believe that this depends both on whether or not the process of digestion was going on at the time, and on whether or not the animals had taken much fluid shortly before their death. Under these different relations I found from 92.342% to 88.684% of water in the serum of portal blood. The serum of hepatic venous blood is always far richer in solid constituents than that of portal blood; in five cases I found the quantity of water in the latter to vary between 89.420%, and 89.298%, a result whose importance in relation to the function of the liver has already been noticed. (See p. 493.)

This leads us to revert to the relation which the amount of water in the serum and in the blood generally bears to the number of blood-corpuscles. It is a striking phenomenon, that *ordinarily* a blood whose serum contains much water, presents few corpuscles; we observe this in blood under various physiological relations (and even in the blood taken from different vessels), but especially in morbid blood; hence, the richer a specimen of blood is in water, so much the more serum or intercellular fluid does it contain: if, however, this is a general rule, it is by no means a law, for we not only meet with exceptions to it, but the most accurate analyses made with special reference to this point fail to establish any constant ratio. Thus, for, instance, in hepatic venous blood there may be from 137 to 351 parts of fresh blood-cells associated with 100 parts of serum containing from 89.3 to 89.4% of water. In morbid blood we still more often meet with similar cases. Hence neither of these properties of the blood depends upon the other, but they are co-ordinate phenomena; that is to say, the conditions which give rise to a diminution of the solid constituents of the serum, generally, at the same time, also occasion a diminution of the colored blood-cells.

It is very difficult to ascertain whether copious draughts of fluid occasion a temporary augmentation of the water in the serum, in consequence of the rapidity with which an excess of water is removed from the blood. Schultz<sup>1</sup> thought that he had convinced himself by direct experiments on oxen, that the blood presented a relative augmentation of water after the copious use of that fluid; Denis, on the other hand, denies that this is the case, at all events in man. But that the solid constituents of the serum should suffer diminution in the absence of proper nourishment, and that there should thence be an augmentation of water, is only what might be expected; and is confirmed by all the investigations that have been made either with healthy or diseased blood, when the persons from whom it was taken had been deprived for a long time of all nutriment, or had been only poorly and scantily fed.

Since in the great majority of *diseases* comparatively little food is taken in consequence of the loss of appetite or the prescription of low diet, and the resorption of nutriment only proceeds imperfectly, or finally essentially nutritious matters are lost by profuse excretions or by copious losses of the juices (as for instance, repeated bloodlettings), it follows, that in consequence of the imperfect restitution of the substances which have been normally or abnormally lost, the blood must become poor in solid constituents. Hence the analyses of the blood in most diseases

<sup>1</sup> Hufeland's Journ. 1838. H. 4, S. 291.

show that it is specifically lighter, that is to say, poorer in solid constituents, than normal blood. This poorness of the blood in solid constituents is not, as a general rule, associated with a diminution of the collective mass or volume of the blood circulating in the vessels; for in our consideration of the mechanical metamorphosis of matter, we shall be led to the result, that the blood has a constant tendency to retain its original volume, so long as the whole mechanism is not disturbed. Hence if solid substances are abstracted from the blood in disease, and are not again replaced, this fluid not only appears watery, in consequence of its containing less solid constituents, but also from its having taken up more water than it contains in the normal state. In such cases the quantity of water is not only relatively, but absolutely increased in the blood. Even in the beginning of most diseases, especially those of an acute character, we find the blood more watery than usual, except during the first ten days of typhus, during cholera, and scarlatina and measles in their first stages; although not unfrequently we find the serum denser and richer in solid constituents than the normal fluid, or at all events, as dense and rich. Hence, it must be concluded that, immediately after the primary invasion of certain diseases, the blood-corpuscles are destroyed in large numbers, or at all events are not renewed in sufficient quantities, and that their products of metamorphosis are retained for some time in the serum, and thus increase its solid constituents, or at all events balance its loss. In the further course of acute diseases (with the exception of cholera), the solid constituents of the serum are always diminished, and its specific gravity falls more or less below the normal standard. The only exceptions to this rule occur in the case of acute articular rheumatism, erysipelas, and puerperal peritonitis; in these diseases there is an extraordinary diminution of the blood-corpuscles, so that the whole blood assumes an abnormally watery appearance, while the serum is denser and contains more solid constituents than in the normal state.

There are certain chronic conditions to which we have applied the names of *anæmia* and *hydræmia*, and which are consequences of severe acute diseases, and especially of such as are associated with considerable losses of the juices, colliquative discharges, or thoroughly destroyed nutrition. The ideas which we are in the habit of connecting with these names are often not sufficiently distinctive. We are accustomed to associate the form of disease which we name chlorosis with both these states, and especially with anæmia. But if, by the term anæmia, we understand an absolute diminution of the blood and of its solid constituents, chlorosis does not fall within the conditions of anæmia; and independently of pathological grounds, the chemical composition of the blood is opposed to this view, for in chlorosis, neither the whole volume of the blood nor the amount of solid constituents in the serum is diminished, but only the number of the blood-corpuscles. Becquerel and Rodier<sup>1</sup> have recently examined the serum with much care in various diseases, and have found that the serum of chlorotic patients presents a perfectly normal constitution. If plethora actually depended on an absolute increase of the blood circulating in the vessels, the not very unfrequent occurrence of plethora in chlorosis would also stand opposed to our attaching identical ideas to the terms chlorosis and anæmia. There is no scientifically ac-

<sup>1</sup> Gaz. de Paris. No. 33 et 36, 1846.



curate proof that there is an actual diminution of the whole mass of the blood, and no such conclusion can be drawn from the appearances after death; hence, if a true diminution of the blood does not exist, our idea of anæmia would entirely coincide with that of hydræmia; and in point of fact, it is in most cases mistaken at the bedside, and confounded with hydræmia. The causes of hydræmia, that is to say of a great excess of water in the blood, and especially in the serum, are sufficiently obvious from the preceding observations. Hydræmia, like dropsy, is only the consequence of an abnormal state of certain organs, for the one necessarily follows the other, each being dependent on purely physical laws; if the blood becomes more watery, the albumen more readily transudes through the capillaries of this or that organ, especially where the motion of the blood is somewhat impeded, and hence the frequency of œdema of the feet; if albumen passes away with the urine, the blood becomes poorer in solid constituents, and the serum more readily transudes; hence, dropsy is a constant attendant on Bright's disease. If, however, dropsy appear sooner than hydræmia, the latter must be the necessary consequence of the former, if abundant transudations of albumen render the blood more watery, without this condition being counteracted by a sufficient renewal of nutriment from without. (C. Schmidt.<sup>1</sup>)

A decided and absolute *diminution of the water* in the serum, and in the blood generally, is in reality only observed in cholera; on this point all observers concur: the watery character of the dejections in cholera, which often contain only from 0·3 to 0·5% of solid constituents, afford a ready explanation of this peculiarity.

In addition to the diseases already mentioned, viz., acute articular rheumatism, puerperal peritonitis and erysipelas, there is also a diminution of the water in the serum, although only a relative one, in chronic diseases of the heart. If, however, symptoms of dropsy have already supervened, we always find that the serum contains an abnormal excess of water.

Before leaving this subject we must remark, that in addition to the proposition that the water of the blood always stands in an inverse relation to the blood-corpuscles, we have also established the aphorism, that the quantity of water in the blood is always proportional to its quantity of fibrin. We must, however, remark that this statement must not be taken literally, that is to say, in a mathematical sense, for we are unable to deduce any formula expressing such a relation. On instituting a comparison between the most accurate analyses which we possess, we just as often find a great augmentation of the fibrin as a diminution of the solid constituents of the blood and of the serum, and the latter are often far more diminished, than the fibrin is increased. Hence it is impossible to refer the augmentation of the fibrin in inflammation, in a direct manner, to the diminution of the albumen, that is to say, to explain the augmentation of the fibrin by a too early metamorphosis of the albumen into this substance, as some have attempted to do. All that we are justified in asserting is this: in those physiological and pathological conditions which are accompanied by a greater or smaller augmentation of the fibrin, we are in the habit of simultaneously observing a diminution of the colored blood-corpuscles, and a greater or less augmentation of the

<sup>1</sup> Charakteristik der Cholera. S. 116-151.

water of the blood, but by no means always of that of the serum; for, to take an example, in acute articular rheumatism, a disease in which the fibrin is often very much increased, we find, on the contrary, the quantity of water in the blood diminished, relatively to the quantity of the solid constituents of the serum; in hydræmia the quantity of water in the serum is extraordinarily increased, while the fibrin scarcely exceeds the normal limits.

We now proceed to the consideration of the *albumen*, of whose occurrence and relations in the blood we have already treated generally (see p. 307).

The amount of albumen in the serum generally rises and falls with that of the other solid constituents; unfortunately, however, most investigations of the blood are limited to the mere determination of the solid residue of the serum, so that we have often no means of determining the ratio in which the latter and the albumen stand to each other: indeed no true conclusion can be drawn from most analyses of morbid blood (previously to those of Scherer and C. Schmidt), not merely because the mode of determining the albumen was unsuitable, but also because we paid too little attention to, or were unable accurately to investigate, the relation of the intercellular fluid to the blood-cells. In order to draw a scientific conclusion from such investigations, it is by no means sufficient to recognize an absolute or a relative augmentation or diminution; it is a much more important point to determine specially in relation to which constituents of the blood the albumen has been increased or diminished; it is not till these highly important relations are followed out in detail, that we can arrive at any inductive conclusions regarding the nature of the pathological changes. Such a general study of the quantitative relations of the albumen in diseased blood, is the means by which we may hope to attain to a true humoral pathology; for, doubtless, all the metamorphoses in the blood proceed from the albumen. We must bear in mind the numerous conditions by which the quantity of albumen in the blood may be changed; this may be effected, not merely by augmentation or diminution of the serum or of the water, but also of salts or extractive matters, by absorption of albumen from the other juices or its loss by exudations or copious excretions, by rich and abundant nutriment, &c. A glance at merely those analyses in which the albumen of the blood has been actually determined by a good method, will indicate the difficulties of attempting to answer such questions.

The quantity of albumen in venous blood increases considerably during digestion.

F. C. Schmid found on an average 6·68% of albumen in the serum from the jugular veins of horses which had been starved for a long time before they were killed; while in the corresponding serum, when the animals had been fed shortly before their death, he found 9·08%.

There is less albumen contained in *arterial* than in *venous* blood, as was discovered long ago by F. Simon. In the serum of the venous blood of the horse I found 11·428%, and in that of the arterial blood 9·217% of albumen. In the residue of the serum of the venous blood there were, however, 15·3 parts of extractive matters and salts to 100 of albumen; while in that of the arterial blood there were 15·7 parts of extractive matters to 100 of albumen.

The serum of *portal blood* is regarded as poorer in albumen than that of the jugular veins; Schmid found on an average 5.19% of albumen in this serum when obtained from fasting horses, and 6.71% when they had been well fed; in horses which had been fed from 5 to 10 hours previously to their being killed, I found from 6.015 to 6.997% of albumen. In the solid residue of the portal serum I found that the albumen stood to the other constituents in the ratio of 100 to 22.5, the horse having been killed five hours after feeding.

The albumen in the *serum of the hepatic venous blood* of horses which were killed from 5 to 10 hours after feeding only varied between 10.487 and 10.702%; hence the serum of the blood of the hepatic veins is far richer in albumen than that of the portal or jugular veins; but if we compare the other solid constituents of the serum with the albumen, we find a diminution of the albumen in the serum of the hepatic veins, as contrasted with that of the portal vein; for while I found the ratio of the albumen to the other solid constituents to be 100 to 22.5 in the serum of the portal blood, it was as 100 to 38.4 in that of the hepatic venous blood. That the albumen in the blood of the hepatic veins is not merely relatively, but also absolutely diminished, is moreover obvious from the composition of the collective blood; in the portal blood we find far more serum than in the blood of the hepatic veins; so that on an average I found that the albumen of the portal blood was to that of hepatic venous blood in the ratio of 3 : 2.

When the intercellular fluid of 1000 parts of portal blood contained 24.453 parts of albumen, 16.553 parts were found in the intercellular fluid of an equal portion of hepatic venous blood; hence the albumen in the two intercellular fluids was in the ratio of 100 : 67.7; in another case the ratio was as 29.606 : 19.806, or as 100 : 66.9; and in a third case (10 hours after feeding) as 44.330 : 32.447, or as 100 : 73.1. Hence, from these numbers, we cannot entertain a doubt that on an average 30.2% of the albumen conveyed to the liver is converted in this organ into other substances, and is probably for the most part applied to the formation of cells.

The reason why Simon<sup>1</sup> found so few blood-corpuscles in the blood of the hepatic veins, is entirely dependent on the analytical method which he employed.

The amount of albumen has been found to be diminished in the following *diseases*: in simple ephemeral and remittent fevers (only slightly diminished), in severe inflammations, in the later stage of typhus (Becquerel and Rodier), in scurvy (where, as is shown by Andral and Gavarret, Becquerel and Rodier, and Favre,<sup>2</sup> it is considerably diminished), in malaria (Salvagnoli and Gozzi<sup>3</sup>), in puerperal fever (Scherer<sup>4</sup>), in dysentery (Leonard and Folley,<sup>5</sup> and C. Schmidt), in Bright's disease, and in dropsy from various organic changes (as was asserted by the older observers, and accurately demonstrated by C. Schmidt). The quantity of albumen in the serum has been found to be increased in intermittent

<sup>1</sup> Journ. f. prakt. Ch. Bd. 22, S. 118.

<sup>2</sup> Compt. rend. T. 25, p. 1136.

<sup>3</sup> Gaz. de Milano. No. 30, 1843.

<sup>4</sup> Untersuchungen, &c. S. 74-69.

<sup>5</sup> Rec. des Mem. de Chim. et de Pharm. milit. T. 60, 1846.

fevers (Becquerel and Rodier), after drastic purgatives, and in cholera (C. Schmidt).

Little importance has generally been attached to the *quantity of fat* in the serum, and we possess very little positive knowledge regarding the quantitative relations of this substance in different physiological and pathological conditions. In most cases in which a determination of the fat has been attempted, this determination has had reference to the blood collectively, so that we have comparatively little information regarding its distribution between the blood-cells and the serum.

It appears from the experiments of Simon, Nasse, Becquerel, and others, that in normal blood-serum the fat ranges from 0.2 to 2.22% of the solid residue.

For further information regarding the quantity of fat contained in the blood generally, we must refer to p. 224.

Although it would appear from the experiments of Boussingault, to which we have already referred, that the use of fat (taken as food) does not induce any augmentation of the fat in the blood, yet nutrition is not without influence on this constituent of the circulating fluid; for during the progress of the digestive process, not only have the chyle and the portal blood been found richer in fat, but sometimes also the serum of the blood generally has been actually observed to be rendered turbid by the presence of this substance (Thomson).<sup>1</sup> Schmid, moreover, found that the serum of horses that had been recently fed contained almost twice as much fat as that of horses which had been kept fasting.

A horse on which I was experimenting was fed for three days, entirely on starch-balls. Immediately before and after this course of diet I abstracted and analyzed the blood from the carotid artery and the jugular vein. The result of this investigation, in reference to the amount of fat, will probably be best shown by the following tabular arrangement.

THE QUANTITY OF FAT

		Before this food.	After this food.
Clot, .	{ From the carotid artery, . . . . .	1.996	1.665
	{ From the jugular vein, . . . . .	2.924	1.366
Serum, .	{ From the carotid artery, . . . . .	2.479	1.465
	{ From the jugular vein, . . . . .	2.984	2.226

This experiment throws light not merely on the constant difference between arterial and venous blood, but also on the influence of an imperfect nutrition—as that of an exclusive starch-diet—on the diminution of the fat in the blood. The number representing the amount of fat contained in the venous clot after the use of the starch, may perhaps be influenced by an error of observation.

The blood of women is, according to Becquerel, generally somewhat richer in fat than that of men.

The serum of *arterial* blood contains less fat than that of *venous* blood: in this respect my results coincide with those of Simon; in the arterial serum of a horse I found 0.264% of fat, which amounted to 2.479% of the solid residue; while in the venous serum I found 0.393%, or 2.984% of the solid residue. In the *serum from the jugular veins* of

<sup>1</sup> Phil. Mag. 3d Series, Vol. 26, pp. 322 and 418.

starved horses, Schmid found that the fat averaged only 0.07% (or 0.93% of the solid residue), while in horses that had been well fed it amounted to 0.13% (or 1.14% of the solid residue).

The difference between the results of my experiments and those of Schmid may appear striking; I must, however, remark that the blood of the horse whose arterial and venous blood were examined before and after the three days' exclusive feeding on starch, contained more fat than that of any other horse I ever met with; this also throws some light upon the numbers (quoted in the next paragraph) which I obtained in a comparative determination of the fat in the portal and the hepatic venous blood, and which are singularly small, although these kinds of blood usually contain more fat than ordinary arterial or venous blood. The blood-cells of this horse did not contain any corresponding augmentation of fat (as may be seen from the previously quoted numbers), so that the great abundance of fat which was presented both by the venous and arterial blood of this horse was entirely limited to the serum. I do not find it recorded in my note-book that the serum was turbid, or that fat-globules were perceived under the microscope.

The serum of *portal blood* is, according to Schultz and Simon, far richer in fat than that of jugular venous blood: in the portal serum of fasting horses, Schmid found on an average 0.10% of fat (or 1.36% of the solid residue), and in that of well-fed horses 0.21% (or 2.06% of the solid residue); I found on an average 0.2843% of fat (or 3.645% of the solid residue) in the portal serum of horses which had been fed from 5 to 10 hours previously.

The serum of the *blood of the hepatic veins* contains far less fat than that of the portal blood, but far more than that of the jugular veins; on an average I found it to contain 0.2722% of fat, or 2.568% of the solid residue.

It is scarcely necessary to remark, that on instituting a comparison of the whole blood (serum + blood-cells + fibrin), the difference which these two kinds of blood present in their amount of fat is far more obvious, because portal blood contains a preponderating quantity of serum, and the hepatic venous blood a comparatively small quantity. The numbers representing the relative amounts of fat are given in p. 481.

The most careful investigations regarding the quantity of fat contained in the serum in different diseases have been instituted by Becquerel and Rodier; from their researches it follows, that almost from the beginning of every acute disease there is an augmentation of the fats in the blood, and especially of the cholesterin. In chronic diseases the fats and principally the cholesterin are especially increased in hepatic affections, as, for instance, icterus and cirrhosis, as well as in Bright's disease, tuberculosis, and cholera.

In the *blood of animals* the quantity of fat appears to be very variable under apparently similar relations; at all events, one and the same observer (as, for instance, Nasse) has found very different quantities of fat in the blood of the same species of animals. This subject has been already noticed in p. 224.

Nasse<sup>1</sup> found the smallest quantity of fat in the blood of goats and

<sup>1</sup> Journ. f. prakt. Ch. Bd. 18, S. 146.

sheep, rather more in that of horses, and still more in that of dogs: the blood of the pig, however, contained no more than that of the dog. While the blood of puppies contained more fat than that of adult dogs, the blood of calves, on the other hand, contained less fat than that of oxen.

Few chemists have extended their inquiries to the determination of the quantity of the *extractive matters* contained in the serum; at all events, they are always determined in association with the salts; the number representing them might certainly be calculated from many analyses, if we did not fear, on the one hand, by including the loss incurred in the entire process, to obtain too high a number, or on the other hand, by imperfect drying, to get by far too low a number. But even when the quantity of the extractive matters has been directly determined, I find from my own investigations, and those of others, that their number is liable to great fluctuations, ranging from 0.25 to 0.42%. When we consider how many things are vaguely included in extractive matters, and how these latter are augmented by the products both of progressive and regressive metamorphosis, we need no longer wonder at these fluctuations.

Nasse has found more *extractive matters* in the blood of *children* and *young animals* than in that of the adult species; the largest quantity was found in human blood, rather less in that of horses, and a much smaller amount in that of oxen.

From the few analyses which I have made with horses' blood, I have been led to the conclusion that more extractive matter is contained in *arterial* than in *venous* blood; while the solid constituents of venous serum contained on an average 3.617% of extractive matters, those of the arterial serum contained 5.374%.

The serum of *portal blood* contains more extractive matters (always determined as free from salts, by the incineration of the ethereal extract freed from fat by water, and of the alcoholic and aqueous extracts) than that of the jugular venous blood; the serum of the *blood of the hepatic veins* contains, however, the largest quantity of extractive matters. In horses which had been fed (from 5 to 10 hours) previously, I found on an average 7.442% of extractive matters (freed from salts) in the solid residue of the serum of the portal blood, and a larger quantity, namely, 10% when the animals had fasted for 24 hours: but from the blood of the hepatic veins I constantly found more than 18% (from 18.1 to 18.5%).

Amongst the *diseases* in which the extractive matters are increased, we may especially notice puerperal fever (Scherer) and scurvy.

For the quantitative determination of the *salts* contained in the serum, it is above all things necessary that we should accurately know the ratio in which the number representing the mineral substances obtained by incineration stands to the number representing the salts which exist pre-formed in the blood, and the manner in which the acids and bases of the ash are grouped in the fresh serum; we know, however, from what has been previously stated, that we too often find great differences in the constitution of the ash, which depend upon the methods we may have adopted for the carbonization and incineration of animal substances. Hence it follows that, notwithstanding the careful labor which so many

inquirers have devoted to the determination of the saline constituents of the blood, the results in question present little uniformity, or, at all events, are of such a character as to preclude us from basing any conclusions on them.

From the best analyses it would seem that the ash of the serum is composed much in the following manner :

Chloride of sodium,	.	.	.	.	.	.	.	61.087
Chloride of potassium,	.	.	.	.	.	.	.	4.054
Carbonate of soda,	.	.	.	.	.	.	.	28.880
Phosphate of soda (2 NaO,PO <sub>5</sub> ),	.	.	.	.	.	.	.	3.195
Sulphate of potash,	.	.	.	.	.	.	.	2.784
								<hr/>
								100.000

The serum of men's blood contains generally rather a larger amount of salts than that of women's blood; the former containing on an average 8.8%, and the latter 8.1%; but the limits between which the amount of salts in the serum of both sexes in the normal state may fluctuate, are tolerably extensive.

According to Nasse and Poggiale,<sup>1</sup> there is a larger amount of salts in the serum of adult men and animals, than in that of children and young animals.

It would appear from the investigations of Nasse and Poggiale, that there is no connection between the saline constituents in the blood of an animal, and the nature of its food; according to these chemists, the blood of cats, goats, sheep, and calves, contains the most salts, then follows the blood of birds, and then that of men, and swine; whilst the blood of dogs and rabbits contains the least.

Nasse found most *alkaline phosphates* in the blood-ash of swine, geese, and hens, and least in that of goats and sheep; he found most *sulphate of soda* in that of sheep, and least in that of hens and geese; most *alkaline carbonates* in that of sheep, and least in that of geese and hens; and most *alkaline chlorides* in that of goats and hens, and least in that of rabbits.

Moreover, the serum of the blood of *different vessels* contains different quantities of salts; from my own investigations and those of Nasse, it appears that arterial serum contains rather more salts than venous serum. Schultz, Simon, and Schmid, found far more salts in the blood of the portal than in that of the jugular vein. (Schmid found at least half as much again.) Moreover, the serum of portal blood contains far more salts than that of hepatic venous blood; in horses we find on an average 0.850% (or 10% of the solid residue) in the former, and only 0.725% (or 7% of the solid residue) in the latter. If to this we add that there is far less serum in the blood of the hepatic veins than in that of the portal vein, it is obvious that the blood of the latter is far richer in salts than that of the former.

By the prolonged use of food rich in common salt, the blood becomes richer in saline constituents, and especially in chloride of sodium (Poggiale and Plouviez).<sup>2</sup>

Zimmermann<sup>3</sup> has found in five experiments made on men, and one

<sup>1</sup> Compt. rend. T. 25, pp. 109-113.

<sup>2</sup> Ibid.

<sup>3</sup> Heller's Arch. Bd. 3, S. 522-530.

observation on a horse, that there is always a larger quantity of soluble salts in the last portion of the blood of one and the same venesection, than in the first portion; and that this augmentation is chiefly due to the alkaline chlorides, the other salts being diminished.

In *diseases* the alkaline salts of the blood undergo considerable fluctuations; but on this point most of the blood-analyses hitherto made are very imperfect; this much only is certain, that in severe inflammations these salts are very much diminished, and that in the acute exanthemata and in typhus they are very much increased. Moreover, C. Schmidt has especially noticed that there is a considerable diminution of the soluble salts in the serum of cholera blood, and an augmentation in dysentery, Bright's disease, and all kinds of dropsy and hydræmia. Finally, it has been found by Leonard and Folley, as well as by Salvagnoli and Gozzi, that the salts are often increased to twice their normal quantity in several endemic diseases, namely, dysentery, malaria, the malignant forms of intermittent fever, scurvy, &c.

It would be highly important to know the amount of *gases* contained in the blood in different physiological and pathological conditions; indeed we hold that it is from this point that a rational investigation of the blood should commence, if we wish to take a philosophical view of its general constitution. All conclusions which we think we can draw from blood-analyses, remain mere conjectures so long as each individual case is not tested by an accurate determination of the gases contained in the blood. Any one desirous of instituting a good analysis of the blood, will not fail to find the means of determining quantitatively the gases of the blood in different diseases; if such an analysis be difficult, it is, at all events, not impracticable, unless physicians adhere to what is now regarded the "rational treatment," and abstain altogether from prescribing venesection. At present we have no certain knowledge on the subject, beyond the results quoted in page 570, for which we are chiefly indebted to Magnus.

We have still to notice some of the more uncommon constituents of the blood, or such as occur in mere traces. We have already mentioned that *sugar* is an integral constituent of the serum. In the blood of oxen, C. Schmidt found from 0.0069 p.m. to 0.0074 p.m. of fermentable sugar; in the blood of a dog 0.015 p.m.; and in that of a cat 0.021 p.m. In the serum of portal blood, in the few cases in which I obtained enough to enable me to detect sugar, I found from 0.0038, to 0.0052 p.m., and in the blood of the hepatic veins, from 0.041 to 0.059 p.m.; in the blood of diabetic patients, where its existence had often been demonstrated, I never could find more than 0.047 p.m. of sugar. This is the more striking as von Becker<sup>1</sup> has found, from numerous and variously modified experiments which he instituted in my laboratory, that, at all events in rabbits, sugar cannot be detected in the urine, unless the blood contains as much as 0.5% of that substance. Von Becker has moreover very distinctly shown, by direct experiments, that highly saccharine food exerts an influence on the amount of sugar in the blood. Thus, for, instance, he found that the blood of rabbits which had been solely fed on carrots yielded 0.584% of sugar, whilst there was only

<sup>1</sup> Zeitschr. f. wissensch. Zoologie. 1853. Bd. 5, S. 123.



0.109% in the blood of those animals when fed upon oats, and only 0.045% in their blood when they had fasted 24 hours. As much as 1.198% of sugar was found in the blood of a rabbit which had been so abundantly supplied with sugar from time to time during several hours, that some of this substance had even passed into the solid excrements.

It was the more important to show, by direct experiments, that food exerted a decided influence on the amount of sugar in the blood, since O. Funke, Bernard,<sup>1</sup> and myself had failed in detecting sugar in the portal blood. Notwithstanding its improbability, the idea readily suggested itself that all the sugar which was found in the blood originated solely in the liver; and that that which was formed during digestion was further metamorphosed in the intestinal canal. Moreover we learn from a careful clinical observation, that, at all events in diabetic patients, a saccharine food exhibits an influence on the amount of sugar in the urine.

There is considerable difficulty in determining the greatest quantity of sugar which can exist in the blood without inducing saccharine urine, and this difficulty may, perhaps, account for the small quantity of sugar found by myself in the blood of a diabetic patient, when compared with that which was found by von Becker in rabbits in which artificial diabetes had been induced by pricking the floor of the fourth ventricle, and in other rabbits; whose blood had been rich in sugar: hitherto von Becker has found that where the blood contains 0.4% of sugar, no portion of it passes unchanged into the urine, although a decided sugar reaction might be detected in the urine obtained by pressure on the region of the bladder, when its quantity in the blood amounts to 0.6% of this substance. The difference in the nature of the urine in man and these animals may perhaps explain the cause of the high amount of sugar which must be present in the blood of rabbits before it appears in their urine, whilst I could discover so little sugar in the blood of diabetic patients; the alkaline urine of rabbits, as we learn from direct experiments externally to the organism, metamorphoses sugar far more rapidly into acid than human urine; we must, moreover, bear in mind that diabetic urine is so poor in matters exciting fermentation, that it passes very slowly into a state of fermentation, which may perhaps in some measure explain the difference. I have, moreover, long since shown that freshly passed urine does not react on vegetable colors in cases of well-marked diabetes, that it is deficient in several of the ordinary extractive matters of normal urine, and that it only gradually acquires an acid reaction on standing exposed to the open air.

We have already noticed the quantities in which, according to Garrod, *uric acid* occurs in normal and morbid blood.

The amount of *urea* in the blood has not yet been quantitatively determined; if, however, as has been maintained, *urea* can be detected in four ounces of healthy blood, its quantity could certainly be easily determined in morbid blood; but ~~this is not~~ the case.

*Silica* was first discovered by Henneberg in the blood of hens, and was determined quantitatively by Millon.

We have already alluded to the occurrence of *carbonate of ammonia*

<sup>1</sup> De urina diabetica. Diss. inaug. Lips. 1835.

in morbid blood; its quantitative determination is impracticable. We would merely add that it has recently been also found in the blood of cholera patients both by C. Schmidt and by myself. While I could detect uræa in the blood of such cholera patients as succumbed before the occurrence of the group of symptoms to which we apply the term uræmia, I always found the blood ammoniacal, and the gastric mucous membrane in the dead body strongly alkaline as soon as the cerebral symptoms peculiar to uræmia had once set in. Moreover, from the analogous experiments which I have instituted with the blood of Bright's disease and scarlatina, I might have been led to the conclusion, that it is not the presence of uræmia, but of ammonia, in the blood, which occasions the symptoms of uræmia; this view is further supported by the experiments of Bernard and Barreswil,<sup>1</sup> who observed that the deleterious consequences of extirpation of the kidneys did not ensue in the dogs on which they operated, until the gastric juice was secreted with an alkaline reaction.

I have just become acquainted with the interesting experiments of Stannius,<sup>2</sup> who found that after extirpation of the kidneys, and even after the simultaneous injection of uræa, uræa itself could never be found in the secretions, or, at all events, in the gastric or intestinal juice or in the bile, but was detected in the sero-sanguineous exudation in the abdominal cavity; but after the death of the animals, the gastric juice, bile, and all the other secretions, were found to be extremely rich in ammoniacal salts. Stannius has thus adduced the most certain proof, that, at all events, the phenomena of uræmia cannot be dependent on the mere retention of uræa. Stannius, moreover, totally denies the possibility of the transmission of uræa into the gastric juice; while I agree with Marchand,<sup>3</sup> and feel convinced that I have ascertained, beyond doubt, the presence of this substance both in the contents of the stomach and in the vomited matters of a dog whose kidneys had been extirpated.

The *bile-pigment*, *biliary acids*, and *abnormal pigments* which are sometimes found in morbid blood, have not been quantitatively determined.

We have already endeavored, in the above remarks, to review the quantitative relations of the constituents of the blood under their various external and internal conditions, and considered the increase and decrease of each individual component part as far as the investigations hitherto made allowed of the prosecution of such an inquiry, this being the only method by which we could hope to arrive at a more thorough insight into the metamorphoses of the blood, and of animal matter generally. It is obvious that we cannot hope to arrive at any definite conclusions regarding the subject in its general bearings, until we have sufficiently examined its individual features under all their different relations. Indeed, the metamorphosis of matter in the blood is entirely comprised in the different relations into which the constituents of the blood are brought under different conditions either in respect to their quantity or quality. We have, therefore, regarded it as more rational and more favorable to the cause of science, to begin our representation

<sup>1</sup> Arch. gén. de Méd. 4 Sér. T. 18, p. 449.

<sup>2</sup> Arch. f. phys. Heilk. Bd. 9, S. 201-219.

<sup>3</sup> Journ. f. prakt. Ch. Bd. 9, S. 499.

of the constitution of healthy and morbid blood, according to the views laid down in p. 411, with a notice of its constituents—that is to say, to consider the blood according to *chemical categories*. In the meantime, we would hope that a short exposition of the results of the analyses of the blood, which have been conducted with reference both to physiology and pathology, may alike tend to throw light upon the whole subject, and to elucidate many physiological and pathological processes. Risking the charge of repetition, we must observe that we purpose giving a short notice of the differences in the constitution of the blood in different physiological and pathological processes, by which method we hope at once to conform to the ordinary mode of treating the subject, and to satisfy the requirements of the practical physician.

In the first place the composition of the blood varies in the different *sexes*. The blood of women is generally of a somewhat lighter red color than that of men; it is specifically lighter, and evolves a less intense odor of sweat when treated with sulphuric acid (Barruel and C. Schmidt); it also contains more water, both in the human subject and in animals. The number of the blood-corpuscles is in general smaller; but there is no perceptible difference in the quantity of fibrin in the blood of the two sexes; hence the serum of coagulated women's blood preponderates over the clot or the blood-cells more than that of men's blood. The serum of the blood in the two sexes differs less than the whole blood, although it generally contains more water. As the serum preponderates in women's blood, it generally contains more albumen than that of men, which is richer in cruor. A similar relation exists in reference to most other constituents of the serum, as, for instance, the fats and extractive matters; but this is not the case with the salts. If we compare the serum of male with that of female blood, we find a larger quantity of salts in the former; if on the other hand, the collective blood of the sexes be compared, we find most soluble salts in that of women.

*Pregnancy* appears to exert the following action on the blood of women; it is generally darker at this than at other periods; its specific gravity sinks in consequence of its becoming richer in water and considerably poorer in colored blood-corpuscles; the fibrin is relatively increased, which generally causes the blood, in coagulating, to form a very small clot with often a superficial stratum of fibrin. The amount of albumen in the serum is also diminished. We have no certain data regarding the fats and salts.

The blood of *children*, and especially of new-born infants, is distinguished by a greater abundance of solid constituents, more especially of blood-corpuscles and iron, while it is poorer in fibrin. It contains, however, nearly the same quantities of fat and albumen as in adult life, and a much larger proportion of extractive matters, and less salts.

In *advanced life*, and in the female sex after the *cessation of menstruation*, the blood becomes poorer in corpuscles; the serum also loses some of its solid constituents; but the cholesterin appears to be somewhat increased.

On comparing the composition of the blood of the different *vertebrata*, we find in the first place, that amongst *mammalia* the *omnivora* exhibit the greatest number of corpuscles, and hence, also, the largest quantity of iron and of soluble phosphates. Fibrin also occurs in larger quanti-

ties here than in the blood of animals of other dietetic habits. The solid constituents of the serum also preponderate in the blood of these animals. The serum of the omnivora contains less salts than that of many other mammalia.

The blood of the *carnivora* generally contains nearly as many blood-cells as that of the omnivora; there is less fibrin but more fat in the blood of these animals than in that of the herbivora. The quantitative relations of the constituents of the blood vary considerably in the different species belonging to this class. A similar remark may be made regarding the blood of the *herbivora*, which on an average contains fewer blood-corpuscles than that of the carnivora, but the deviations from this rule are as great in the different species of this class as in the carnivora. We may, however, hope that a more careful study of the composition of the blood of these three groups of animals will enable us to detect more definite differences between them.

The blood of *birds* is rich in corpuscles, and stands next in this respect to that of the pig; it contains, however, more fibrin and fat, and less albumen, than that of the mammalia.

In the *cold-blooded vertebrata* the blood is poorer in corpuscles, and richer in water than in the other *vertebrata*.

Although the *mollusca* possess a vascular system, consisting of arteries and veins and an aortic heart, their blood differs very considerably from that of the classes of animals immediately above them; being a white or bluish juice. C. Schmidt<sup>1</sup> found the blood of the pond-mussel (*Anodonta cygnea*) colorless and slightly alkaline; it deposited a pale, fibrinous coagulum, which on evaporation exhibited beautiful crystals resembling Gaylussite, and consisting of carbonate of lime and some carbonate of soda. The albumen was mostly combined with lime. This blood contained only 0.854% of solid constituents, and of these there were 0.033 of a fibrin-like substance, 0.565 of albumen, 0.189 of lime, 0.033 of phosphate of soda, chloride of sodium and sulphate of lime, and 0.034 of phosphate of lime.

E. Harless and v. Bibra<sup>2</sup> investigated the blood of the large Shell-snail (*Helix pomatia*) and that of certain Cephalopods (*Loligo* and *Eleuthero*), as well as of certain Tunicata (as, for instance, of some Ascidians).

The blood of the large Shell-snail contains, according to their investigations, 8.398% of organic, and 6.12% of mineral substances, there being 0.033 of oxide of copper in the latter. This blood is especially distinguished by assuming a blue color on exposure to the air, in consequence of the access of oxygen, and again becoming colorless by the action of carbonic acid. Alcohol yields a colorless coagulum; ammonia removes the blue color, which is restored by neutralization with hydrochloric acid. The blue pigment is precipitated by alum and ammonia, and is entirely destroyed at 50°. The blood of the Ascidians and Cephalopods presents the opposite relations in regard to color to that of the large shell-snail. It is not colored blue either by oxygen or nitrogen, but carbonic acid converts it into an intense blue. Oxygen does not cause the entire dis-

<sup>1</sup> *Zuvergleichenden Physiol.* Mitau, 1846, S. 58-60 [or Taylor's Scientific Memoirs, vol. 5, p. 26].

<sup>2</sup> *Müller's Arch.* 1847. S. 148-157.

appearance of this color; while ether and alcohol instantly communicate a blue color to the originally colorless blood. Bibra found in this blood 4.7% of organic and 2.63% of mineral substances, but no iron, although some copper.

Genth<sup>1</sup> has also recently examined the ash of the blood of *Limulus* Cyclops, which, when fresh, has an azure blue color, and has found in it a considerable quantity of copper with a little iron. In two analyses of the ash of this blood, he found in 100 parts:

Oxide of copper, with traces of oxide of iron, . . . . .	0.297 . . . . .	0.083
Chloride of sodium, . . . . .	72.907 . . . . .	83.507
Phosphoric acid, . . . . .	0.683 . . . . .	0.281

The blood had a specific gravity of 1.0317 and yielded 3.327% of ash.

I have made some experiments<sup>2</sup> on the blood of *insects*, and especially of the lepidoptera in their larva state. On making an incision into the skin of a caterpillar, on the abdomen, a transparent, thick, pale yellowish green juice exudes, which under the microscope discloses roundish cells without a distinct nucleus; the cell-walls appearing stippled like those of pus-corpuscles and having a diameter varying from  $\frac{1}{350}$ ''' to  $\frac{1}{200}$ '''. Dilute acetic acid does not change the cells, but the concentrated acid dissolves them. Caustic alkalies cause them to conglomerate into masses like most cells and even the yeast-globules, making them appear somewhat relaxed in texture, distorted and granular, so that they resemble granular cells. Hydrochlorate of ammonia does not change them. Besides these cells, we very frequently observe large roundish oval cells, having a distinct nucleus, and not unlike many of the pavement epithelium cells. These are not changed by acetic acid or the caustic alkalies. More rarely there occur pyriform, or spindle-shaped, and other irregularly formed cells. Fat-globules are always present in this fluid; they might be referred to the fat surrounding the stomach; if they did not likewise occur in the fluid of the dorsal vessels.

The intercellular fluid of the blood of insects assumes a dark brownish-green, or even black shade, when exposed to the air, and becomes turbid from the deposition of very fine molecular granules. It has a faint alkaline reaction, speedily developes ammonia on exposure to the air, and coagulates on being boiled, as well as on the addition of mineral acids or of a watery solution of iodine, into a thick white mass, without any separation of serum. It is also rendered turbid by water, and then resembles under the microscope a finely granular mass in which long threads are plainly discernible. Hydrochlorate of ammonia does not remove the turbidity, and the caustic alkalies or acetic acid remove it only slightly. Dilute acetic acid causes the fluid to gelatinize, and removes the blackish-green color, if it had previously been induced by exposure to the air. The caustic alkalies also convert the clear fluid into a colorless, tenacious jelly. Sugar may sometimes, but not always, be detected in this fluid. As caterpillars generate a larger quantity of fat within a short period than any other animals, their blood is also the richest in fat; amounting in one experiment to 27.5% of the solid resi-

<sup>1</sup> Keller u. Tiedemann's Nordam. Monatsschr. Bd. 3, S. 438-441.

<sup>2</sup> Goschen's Jahresb. Bd. 2, S. 19.

due. The fluid of the dorsal vessels in insects does not appear to differ essentially from the above-described juice, containing precisely the same elements, with the exception of those nucleated cells which resist the action of acetic acid and the caustic alkalies.

The *blood of the arteries* differs from that of the *veins* in containing a smaller quantity of the solid constituents belonging to the blood-cells, which however contain relatively more hæmatin and salts than the cells of venous blood, but far less fat. The intercellular fluid of the arterial blood is richer in fibrin than that of venous blood. The serum of the former contains somewhat more water, and consequently less albumen; for if we compare the solid constituents of the serum of both kinds of blood in regard to their quantity of albumen, we shall find an equal amount of this substance in each. The case is different with the fats, extractive matters, and salts; for the first are considerably diminished in the arterial fluid serum, and even in its solid residue; and while the salts are but slightly augmented, the extractive matters are considerably increased in quantity. The arterial blood moreover contains relatively more free oxygen than the venous blood.

The *portal blood* differs in constitution according to the different stages of the digestive process; *during digestion*, when drink, as well as food has been partaken of, it is rich in water and intercellular fluid; the number of blood-corpuscles is therefore small, the fibrin is slightly, and the fat very considerably augmented, while the albumen, extractive matters, and salts are moderately increased. The fibrin during the digestion remains the same as in the other vessels, but after the completion of that process it can readily be torn, and forms only a loose diffuent clot.

*Compared with the blood of the jugular veins*, portal blood is poor in cells as well as in solid constituents generally; these cells are partly flocculent, are easily distorted, and soon become jagged after their removal from the body. They are richer in hæmatin and poorer in globulin than the cells of the blood of the jugular veins, but contain twice as much fat. The intercellular fluid contains a fatty fibrin, which, however, is inferior in quantity to that in the blood of the jugular veins. The serum contains on an average less solid constituents generally (especially albumen), but more fat, extractive matters, and salts. Biliary substances have not been shown to exist in portal blood, and sugar only seldom occurs.

The *blood of the hepatic veins* differs in constitution from that of any other vessels. *Compared with portal blood*, it is poor in water; for if we assume the solid constituents of the two kinds of blood to be equal, the amount of water in the portal blood will be to that in the hepatic venous blood during digestion, when little fluid has been taken, as 4 : 3, and after the completion of digestion not unfrequently as 12 : 5. The clot of hepatic venous blood is voluminous, and readily falls to pieces. While 100 parts of portal blood yield 34 of serum, 100 parts of hepatic venous blood yield only 15 of serum. Hepatic venous blood is far richer than portal blood both in colored and colorless cells, the latter presenting every variety of size and form, and the former exhibiting heaps of a distinct purplish-red color. Their cell-walls are less easily destroyed than those of the blood of other vessels. While in the corresponding portal

blood there are 141 parts of moist blood-cells for every 100 parts of intercellular fluid, there are in hepatic venous blood 317 parts blood-cells for 100 parts of the intercellular fluid. The cells of the latter blood are poorer in fat and salts; especially poor in hæmatin, or at least in iron, but somewhat richer in extractive matters. These cells have a greater specific gravity than those of portal blood (notwithstanding the diminished quantity of iron). On comparing the specific gravity of both kinds of blood with that of the serum, we find that the cells are lighter in relation to the serum in the blood of the hepatic veins than in that of the portal vein. The intercellular fluid of the former is far denser than that of the latter; it also contains a much larger quantity of solid constituents; but, on the other hand, it is either wholly deficient in fibrin, or only contains it in scarcely perceptible traces. While in the portal serum there are 8.4 parts of solid substances to 100 parts of water, there are 11.8 parts of solid matters to 100 parts of water in the serum of hepatic venous blood. When we compare the solid constituents in the serum of both kinds of blood, we find that hepatic venous blood contains less albumen and fat, and a much smaller quantity of salts; while the extractive matters, including sugar, are considerably increased. In the solid residue of the hepatic venous blood of horses, I found in three determinations (in which the alcoholic extract was excited to fermentation by means of yeast, and the sugar,  $C_{12}H_{12}O_{12}$ , was calculated from the developed carbonic acid) that the sugar was respectively 0.635, 0.893, and 0.776%; whilst in the residue of the corresponding portal blood I only once succeeded in detecting sugar, and then it only amounted to 0.055%.

The *blood of the splenic vein*, which has only been chemically examined, and compared with that of the jugular vein in horses and dogs by Bécclard,<sup>1</sup> contains more water than the last-named kind of blood. The mean of 14 investigations in the case of dogs was 77.815%, the extremes being 74.630 and 82.681%. The corresponding jugular venous blood contained, on an average, 1.608% less water than the blood of the splenic vein. In two parallel investigations of horses' blood, the latter kind contained from 0.4 to 0.5% more water than the jugular venous blood. The blood-corpuscles are somewhat diminished, but the fibrin and the residue of the serum somewhat increased, in the blood of the splenic vein. Ecker<sup>2</sup> also found in the latter blood the cells containing corpuscles, discovered by Kölliker in the splenic juice. This was especially the case in the splenic venous blood of horses. From 1 to 5 corpuscles, or small yellow granules, were found enclosed in one capsule.

Funke's very carefully conducted examination of the blood of the splenic vein in horses does not, unfortunately, either confirm or refute Bécclard's conclusions, while my own experiments on the arterial blood of the same animals (from which the blood of the splenic vein had been taken) exhibited such different results, that no general deductions could be obtained in reference to any one point. The juice expressed from the spleen which J. Scherer<sup>3</sup> analyzed consisted principally of blood,

<sup>1</sup> Gazette Méd. 1848. No. 4, p. 22, Janv.

<sup>2</sup> Handwörterb. de Physiol. Bd. 4, S. 146.

<sup>3</sup> Verhandl. d. phys. med. Ges. zu Würzburg. Bd. 2, S. 323.

yielded by the capillaries of the spleen. Scherer found that it contained in addition to albuminous matters and salts, lienine, hypoxanthine, two different kinds of ferruginous pigments, a large amount of free iron not combined with pigments, and acetic, formic, and lactic acids.

This investigation of Funke affords, at all events, a proof that the greatest caution is necessary in deducing conclusions from individual analyses and investigations of individual fluids, without reference to the simultaneous constitution of the other animal juices. Many ingenious conclusions would no doubt have been deduced from analyses of the blood of the splenic vein, if the arterial blood had not been simultaneously compared with it.

The *menstrual blood* contains no fibrin, as was shown by Jul. Vogel in the case of a person suffering from prolapsus uteri, and has been recently confirmed by C. Schmidt.<sup>2</sup> It yields a colorless but distinctly alkaline serum and a red deposit of blood-corpuscles; these are interspersed with numerous colorless cells, but there is no trace of the so-called fibrinous flakes. It contains about 16% of solid constituents.

Henle believes that the only reason that the menstrual blood does not coagulate, is because each individual drop forms a distinct coagulum, and that consequently the sum of the drops must always constitute a tolerably fluid mass; but when examined under the microscope, menstrual blood does not exhibit any coagulated substance near or among the corpuscles. On the other hand, E. H. Weber found coagulated blood upon the mucous membrane of the uterus of a young girl, who had killed herself during the period of menstruation.

The blood of the *placental vessels* contains, according to Stas,<sup>3</sup> little albumen and fibrin, whose place is, however, supplied by a large amount of a substance which he calls casein (see p. 342). Stas also believes that he has found urea in this blood.

During *digestion*, the blood becomes richer in solid constituents, this increase extending with tolerable uniformity to the blood-cells and the plasma. The former gain in solid constituents, while they experience a relative loss of hæmatin (F. C. Schmid). The fibrin of the intercellular fluid is scarcely perceptibly increased, but it coagulates more slowly, and therefore more readily forms a crust upon the clot. Lastly, it is richer in fat than the fibrin obtained from the blood of fasting animals; the serum is denser, sometimes even exhibiting a milky turbidity from fat-globules and colorless blood-cells. It also presents a tolerably uniform proportional augmentation of fat, albumen, extractive substances, and salts.

*Prolonged fasting and extensive losses of blood or of the other juices* exert an action on the constitution of the blood precisely analogous to that of those substances which interfere with digestion or resorption and the formation of blood; as, for instance, many metallic salts, and especially preparations of lead, acids, &c. In these conditions, the number of the corpuscles diminishes in various degrees, while the plasma be-

<sup>1</sup> Wagner's Lehrb. d. Physiol. 2 Aufl. S. 230.

<sup>2</sup> Diagnostik verdächtiger Flecke. Mitau u. Leipzig, 1848, S. 8 u. 41.

<sup>3</sup> Compt rend. T. 31, p. 630.



comes more watery (that is to say, poorer in albumen and other organic constituents), but richer in salts. The blood has nearly the same constitution as in anæmic conditions.

C. Schmidt<sup>1</sup> has endeavored by careful and numerous experiments to establish the proposition, that *the loss of albumen in the blood is supplied by a relatively corresponding amount of salts*, as for instance, *chloride of sodium*. Thus we find that, wherever albumen is lost from the blood, either by accidental or intentiona! bloodletting, by morbid exudation from the capillaries of the serous membranes (dropsy), by the action of the kidneys (albuminuria), or by other losses of the juices whose action is manifested by a diminution of albumen in the blood, certain quantities of the albumen lost from the blood are replaced by certain quantities of soluble salts, and here we must bear in mind that the salts are in general accompanied by a definite quantity of water, which differs from the amount associated with the albumen. The experiments of Kierulf<sup>2</sup> have recently furnished a new proof of the correctness of these observations, for he found that after a considerable quantity of water had been injected into the veins, the amount of the salts in the blood was rapidly and permanently increased.

In order to determine the influence exerted on the constitution of the blood by the abstraction of that fluid, numerous experiments have been made by Nasse on healthy animals, and by Becquerel and Rodier, Zimmermann, and others, on persons in disease. The results obtained showed that the specific heat, as well as the specific weight of the blood, was diminished; in color, the blood was more brightly red; it coagulated more rapidly, but there was a less thorough expression of the serum, which exhibited a reddish or whitish turbidity. The red corpuscles, which were much diminished in number, showed a greater tendency to cohere. The colorless cells were increased in number (Nasse, Remak), and the quantity of water was considerably augmented; and at each venesection the blood became poorer in cells than in the solid constituents of the serum. The quantity of the fibrin was scarcely increased in healthy animals, and in disease it is altogether independent of the abstraction of blood. The blood-cells became poorer in globulin, and relatively richer in hæmatin (C. Schmidt).

These facts seem in some degree connected with the differences observed in the constitution of *different portions of blood taken at one and the same venesection* by Prevost and Dumas especially, but also by Becquerel and Rodier, and Zimmermann. After the loss of the first portion of blood (about 100 grammes), the solid constituents are in no case increased in the second portion, but on the contrary they almost always diminish with tolerable uniformity, while a third portion very frequently exhibits an increase of solid constituents when compared with the second (Zimmermann). This diminution of the solid substances depends upon the resorption of fluid, which obviously is owing to the absorption not of pure water, but of lymph, fluid exudations, and parenchymatous juice, which are lighter than the blood. The amount of absorption of water varies, however, very considerably in special cases.

<sup>1</sup> Charakteristik der Cholera, S. 60.

<sup>2</sup> Mitth. d. naturf. Ges. z. Zürich. Juli, 1852.

In Becquerel's experiments the quantity of water increased almost uniformly with each portion of blood, till it attained its maximum in the last that was drawn.

*Inflammatory diseases* constantly induce an increase of fibrin, when the inflammation is accompanied with fever. The number representing the fibrin is in general increased in the largest proportion in acute articular rheumatism and in pneumonia. A considerable increase of fibrin may be induced even where inflammation of a tissue is not very extensively diffused, as, for instance, in erysipelatous inflammations. In each individual disease the quantity of fibrin in the blood increases in proportion to the degree and duration of the inflammation. The increase of this substance is independent of the condition of the patient as to strength, and unconnected with the increase or decrease of the other solid constituents of the blood. Even in the most decided anæmia or hydræmia, the inflammation induces an augmentation of the fibrin. As the blood of persons who have died from acute cerebral diseases has never been found in a state of coagulation, it appears not wholly irrelevant to observe that in meningitis, &c., the blood removed from the living body has been found to be as rich in fibrin as it is in any other form of inflammation.

The number of the red blood-cells is decreased during the febrile inflammatory process, although not to any very great degree, unless the existence of other pathological processes has induced a simultaneous diminution of the whole mass of the blood-cells. In some cases scarcely any diminution of the blood-cells can be observed, although there may be a considerable increase of the fibrin.

The diminution of the solid constituents is in general proportional to the violence of the inflammation, and also to the quantity of exudations thrown off. Where there has been no great amount of exudation, the solid constituents are sometimes found to be augmented rather than diminished (as for instance in bronchitis). The diminution of the solid residue of the serum depends mainly upon the decrease of the albumen; for the salts in the serum are unaltered, and the fats, or rather the cholesterin, may be considerably increased.

We cannot at the present time attempt to decide whether the group of symptoms which accompany most acute diseases, and are designated as *fever*, are characterized by certain constant alterations in the relative quantities of the blood-constituents; but all investigations agree in showing that fever itself exerts neither an increasing or decreasing action on the vacillating amount of fibrin in the blood. The inquiries hitherto made, do not warrant us in deciding whether the admixture of the blood, which Becquerel and Rodier believe they have found to exist during the development of every acute disease, can be regarded as peculiar to fever. According to these authors, the blood presents at this time the following appearances: it is in general somewhat more watery than in its normal state; the corpuscles are slightly diminished in number, while among the fats, the cholesterin and the phosphorized fats are especially increased; the extractive matters and the soluble salts occur in normal quantity, while the phosphates are considerably augmented.

The same inquirers found that the blood-corpuscles, as well as the

fibrin and the soluble salts of the serum, occurred in their normal quantity in simple *ephemeral* and *remittent fevers*, while the albumen was slightly diminished and the cholesterin increased.

In slight *intermittent fevers*, Zimmermann found that the fibrin was only increased in some few cases, being more frequently diminished, but it in general occurred in the normal quantity. Its increase appeared to stand in a direct relation to the duration of the fever. Becquerel and Rodier found the fibrin diminished in most cases of intermittent fever.

In *endemic intermittent fevers*, the blood-corpuscles are seldom diminished to any considerable degree, except in relapses, but are frequently increased in quantity. The fibrin is invariably augmented in inflammatory affections. The constituents of the serum increase when the disease presents an intermittent type, but decrease when the disease is characterized merely by remissions of severity. The diminution in the latter case mainly affects the albumen, while the salts of the serum are constantly augmented in quantity.

In *marsh-fevers* (malaria), the corpuscles are considerably increased (Salvagnoli and Gozzi, Luderer), while the fibrin, albumen, and fats, are proportionally diminished. A large quantity of cholesterin, as well as of bile-pigment, is in general found.

In *cholera* the blood is especially dense and viscid; and while the blood-corpuscles are relatively augmented, they are poorer in salts. The fibrin remains unaltered as to quantity; the serum is denser, poorer in water and salts, but relatively very rich in albumen; it also contains more potash salts and phosphates than normal serum, some urea, and an extractive substance by which urea is rapidly converted into carbonate of ammonia.

In *dysentery* the blood is poor in corpuscles. The fibrin is generally, although not always, somewhat increased. All the solid constituents of the serum are decreased, but especially the albumen. The salts, on the other hand, are considerably increased in quantity.

In *Bright's disease* the blood presents not only a considerable diminution in the number of cells, but likewise a great loss of the constituents of the serum. The cholesterin as well as the salts of the serum are, however, augmented, and the fluid almost always exhibits traces of urea, which in some cases is present in considerable quantity. Such blood contains on an average more fibrin than in the normal state, while it is only in inflammatory affections of the kidneys, that is to say, in its first stage, that there is any great augmentation of fibrin.

The *hydræmic blood* observed in different kinds of dropsy, is a very attenuated, pale, watery fluid; in coagulating it forms a very loose, infiltrated gelatinous clot. Its composition is very similar to that observed in Bright's disease, almost the only point of difference being the absence of urea. According to my experience, at all events, this substance does not occur in hydræmic blood more than in dropsical exudations, unless in those cases in which renal affections are simultaneously present.

If by the term *anæmia* we understand a diminution of the quantity of blood in the vessels (*olichæmia* would, therefore, be a more correct expression, etymologically), we can scarcely assert that the blood exhibits

a perfectly identical or even an analogous composition in all conditions included under this designation, since the composition of the blood must necessarily correspond with the morbid process which preceded the diminution of the blood; for the properties which have commonly been ascribed to anæmic blood belong, properly speaking, to a hydræmic condition. We must, at all events, presume that the blood in anæmia depending upon excessive hemorrhage, differs in composition from that exhibited in the anæmia which arises from large tumors, excessive mental labor, bad food, poisoning, &c. Experience teaches us, moreover, that the anæmia which follows carcinoma, typhus, hemorrhages, and other losses of the juices, may easily pass into hydræmia, whilst in tuberculosis a hydræmic state of the blood is scarcely ever found to occur together with the corresponding serous exudations. Anæmic blood does not therefore indicate the existence of any special admixture of the blood. It is only in respect to the diminution of the colored blood-cells that the composition of this blood corresponds with that exhibited in hydræmic and chlorotic conditions.

In *chlorosis* the blood forms a small solid clot covered with a buffy coat, and floating in a large quantity of clear serum. The corpuscles and the iron are both diminished either in a very small or in an excessive degree, without, however, standing in any definite relation to the intensity of the disease. The quantity of fibrin does not greatly exceed the normal average; the quantity of albumen is only increased relatively to the blood-cells, while the fats and salts remain entirely normal.

In the so-called *plethora*, the blood-corpuscles are always somewhat more numerous; the serum and the fibrin are both nearly normal, and the albumen of the liquor sanguinis rises only slightly above the mean average. Plethora seems to bear the same relation to spinal irritation as anæmia does to chronic spinal affections, the only difference being a greater increase of the solid constituents, and more especially of the blood-corpuscles, in the former.

The blood experiences no changes in *typhus*, which can justify us in terming this disease a dyscrasia. From the 5th to the 8th day, and therefore nearly as long as the continuance of the typhus exanthema, we find that the composition of the blood bears a great similarity to that exhibited in plethora, for the corpuscles are increased, as also are the solid constituents of the serum, and especially the albumen; even the fibrin is generally augmented at this period. From the 9th day of the disease the constitution of the blood assumes a totally different character; for at this period the blood becomes lighter, chiefly owing to a diminution of the corpuscles; the residue of the serum, however, diminishes daily through the entire duration of the disease, with a rapidity proportional to the intensity of the intestinal affection. The salts and extractive matters are relatively increased, rather than absolutely diminished. If typhus be not followed by any of its frequent sequelæ, or by the anæmia accompanying many of the epidemic forms of this disease, there is generally found to be an increase of the solid constituents about the beginning of the fourth or fifth week, which in some cases chiefly affects the blood-corpuscles, in others the solid substances of the serum, while occasionally even the quantity of the fibrin is augmented.

In *acute exanthemata*, there is a diminution of the blood-cells and a corresponding augmentation of the intercellular fluid. The serum is denser than usual, and its salts are far more augmented than the organic substances.

In *puerperal fever*, the blood varies according to the course and character of the morbid process (as indeed we observe in most cases of disease). There is a very considerable diminution of the corpuscles; the fibrin, especially in peritonitis, is much increased, but is soft and gelatinous, and almost always forms a crust. In most cases the solid constituents of the serum are considerably diminished (Scherer, Becquerel and Rodier); but sometimes they are increased (Andral and Gavarret); the extractive matters are considerably increased (Scherer); bile-pigment is occasionally met with (Heller); and not unfrequently free lactic acid (Schärer).

In *pyæmia*, the fibrin is diminished, and the colorless blood-cells augmented; but more than this is not known, as the blood has not been carefully examined in this disease.

In *leucæmia*, which is commonly associated with a considerable enlargement of the spleen, the entire mass of the blood exhibits considerable similarity with the blood of the splenic vein (Virchow, Scherer). The blood from the most different vessels is pale red, often marked with whitish streaks and very rich in colorless blood-corpuscles; within the body it coagulates into gelatinous flakes, but when it coagulates in the air, very little serum separates from it; it exhibits an alkaline reaction, although the fluid which is filtered from the coagulum has an acid reaction; according to Scherer's investigation, this blood contains true glutin, also a body which ranks between glutin and albumen (an albuminous substance containing phosphorus and iron), hypoxanthine, and finally formic, acetic, and lactic acids. In other respects, according to Scherer's analysis,<sup>1</sup> its quantitative composition is nearly the same as that of normal blood in respect to the main constituents, excepting that the iron seems to be present in a somewhat smaller quantity.

The blood has not been examined with accuracy in *scurvy*, and its composition has therefore been deduced principally from physical relations; thus, for instance, its imperfect coagulation led to the conclusion that it exhibited a diminution of fibrin, while other causes led in the same manner to the supposition that there was an augmentation of the salts. The few investigations of scorbutic blood which we possess, give but little idea of the true constitution of this fluid in the condition which we term scurvy. Becquerel and Rodier have been led by their most recent analyses, which, however, are not very conclusive, to adopt the opinion that "the essential anatomical character of scurvy must be sought in an original modification of the fibrin," while they show that there is an increase of the fibrin in the acute form of the idiopathic disease, "depending upon an excess of the soda-salts in the blood."

The admixture of the blood in *tuberculosis* does not seem to differ greatly from the normal condition, for the modifications which it undergoes, appear, as far as our chemical investigations have enabled us to judge, to depend entirely upon the conditions which accompany this dis-

<sup>1</sup> Verh. d. physik-med. Ges. zu Würzburg. Bd. 2, S. 321-325.

ease; thus, in inflammatory affections, the blood presents the same composition as in inflammations, while in those cases in which there is considerable loss of blood from hæmoptysis, or when profusely discharging intestinal ulcers or colliquative sweats are present, all the solid constituents of the blood, excepting the salts, decrease, as do also the blood-cells with even greater rapidity. Dropsy is not often associated with tuberculosis, but when this combination does occur, the blood presents the appearance of hydræmia.

The blood has not yet been very carefully examined in *carcinoma*; it is, however, worthy of notice that Popp, as well as Heller, and recently also v. Gorup-Besanez,<sup>1</sup> have discovered an increase of fibrin in carcinoma, even when unassociated with febrile affections. (It is certainly not shown whether the substance in excess was true fibrin.) The number of the blood-corpuscles is somewhat diminished. When dropsy is associated with cancer, the blood becomes hydræmic. As the solid constituents of the serum are not often abnormally increased, we cannot suppose that there is any serous or albuminous crasis in carcinoma.

Although we should naturally expect to find that the constitution of the blood undergoes a special alteration in *diabetes*, no such change has as yet been discovered; for, excepting its increased quantity of sugar, it presents nearly the same composition as normal blood: It is somewhat more watery, and contains less fibrin, but the blood-cells and solid constituents of the serum are only slightly diminished (v. Gorup-Besanez even found them increased). The serum sometimes exhibits a milky turbidity (Thomson).

The conception or idea of *scrophulosis* is as indefinite as that of *chronic rheumatism* and *arthritis*; and hence no scientific investigation of the blood in those conditions can be entered upon, for the blood must necessarily possess a different constitution when the scrofulous swellings of the cervical glands arise from ulcers on the pharyngeal mucous membrane, and when they depend upon tuberculous deposits. The constitution of the blood cannot be the same when uric-acid concretions are deposited in the joints, and when necrosis, osteoporosis, or osteosclerosis is established in consequence of periostitis. It has been asserted that the blood in scrofula is remarkable for its poverty of cells (Nicholson),<sup>2</sup> and that arthritic blood is distinguished by the presence of uric acid and urea (Garrod).<sup>3</sup>

The immediate effect of the inhalation of ether seems to make the blood richer in water, poorer in blood-corpuscles, and strikingly rich in fat (Lassaigne,<sup>4</sup> v. Gorup-Besanez).<sup>5</sup> According to the numerous investigations of Gorup-Besanez,<sup>6</sup> no distinct relation can be discovered between the *bruit* in the jugular veins and the chemical constitution of the blood. This sound may exist where there is an increase of all, or of some only of the solid constituents of the blood, or where they are diminished, or, finally, where there is a perfectly normal composition of the blood.

<sup>1</sup> Arch. f. physiol. Heilk. Bd. 8, S. 522-525.

<sup>2</sup> Lancet. Nov. 1845, p. 451.

<sup>3</sup> London Med. Gazette. Vol. 31, p. 88.

<sup>4</sup> Gaz. Méd. de Paris. No. 11, 1847.

<sup>5</sup> Arch. f. physiol. Heilk. Bd. 8, S. 515-523.

<sup>6</sup> Ibid. p. 532-543.

The *quantity of blood contained in the living body* has never been accurately determined, for the simple reason that the entire mass of the blood cannot be completely removed from the vessels and weighed; hence the determination can only be made approximately by indirect methods. Herbst endeavored to calculate the quantity of blood in the vessels by the quantity required for the complete injection of the veins and arteries. But all who have made injections, or even carefully examined the injected subject, must feel that the estimate will be very uncertain when based upon such methods. Vogel,<sup>1</sup> Dumas,<sup>2</sup> and Weisz,<sup>3</sup> have proposed but not practised other methods of determination. Valentin<sup>4</sup> suggested the ingenious expedient of abstracting blood from an animal whose weight was known, and after determining the solid constituents, immediately injecting a certain quantity of pure water into the veins, and then again taking blood and examining the solid residue with the greatest care. From the difference in the amount of the solid constituents in the two different kinds of blood, Valentin calculated the ratio of the weight of the whole blood to that of the body in dogs and sheep as  $1:4\frac{1}{2}$  in the former, and  $1:5$  in the latter. This method would afford sufficient accuracy if the walls of the bloodvessels were not more easily permeated by a thin than by a dense plasma,—if the whole mass of the juices in respect to the amount of water did not stand in such a relation to the blood that the state of the latter is almost immediately reflected in them (as indeed we see from the different composition of the separate portions of the blood in one and the same venesection, see p. 632),—if the blood did not continually give off water to the kidneys and other excretory organs,—and lastly, if the vessels were mere water-proof canals, without openings for the escape of the water, and for the importation of solid parts.

The discrepancies in the views of different physiologists in reference to the quantity of blood contained in the body of an adult man, are sufficiently obvious, when we remember that Blumenbach estimated it at 4 or 5 kilogrammes [from 8·5 to 11 pounds], and Reil at fully 20 kilogrammes [or 44 pounds]. In the present day the blood is generally estimated at 10 kilogrammes [or 22 pounds], which is equal to about the 8th part of the weight of the whole body. If I may advance the opinion at which I have arrived from experiments prosecuted on the bodies of two executed criminals, I should estimate the blood in the body of a young man as somewhat below the above quantity, namely, at from about 8 to 8·5 kilogrammes [or from 17·5 to nearly 19 pounds].

My friend, Ed. Weber, determined, with my co-operation, the weights of two criminals both before and after their decapitation. The quantity of the blood which escaped from the body, was determined in the following manner: water was injected into the vessels of the trunk and head, until the fluid escaping from the veins had only a pale-red or yellow color; the quantity of the blood remaining in the body was then calculated, by instituting a comparison between the solid residue of this

<sup>1</sup> Pathol. Anat. des menschl. Körpers. Leipz. 1845, S. 59 [or English translation, p. 84].

<sup>2</sup> Chim. physiol. et méd. Paris, 1848, p. 326.

<sup>3</sup> Zeitsch. d. k. k. Gesellsch. d. Aertze. Dec. 1847, S. 203-229.

<sup>4</sup> Repert. der Physiol. Bd. 3, S. 281-293.

pale-red aqueous fluid, and that of the blood which first escaped. By way of illustration, I subjoin the results yielded by one of the experiments: the living body of one of the criminals weighed 60140 grammes, and the same body after the decapitation 54600 grammes; consequently, 5540 grammes of blood had escaped. 28·560 grammes of this blood yielded 5·36 grammes of solid residue; 60·5 grammes of sanguineous water collected after the injection, contained 3·724 grammes of solid substances. 6050 grammes of the sanguineous water that returned from the veins were collected, and these contained 37·24 grammes of solid residue, which corresponds to 1980 grammes of blood; consequently, the body contained 7520 grammes of blood (5540 escaping in the act of decapitation, and 1980 remaining in the body); hence, the weight of the whole of the blood was to that of the body nearly in the ratio of 1:8. The other experiment yielded a precisely similar result.

We have no intention of asserting that such experiments as these possess extreme accuracy, but they appear to us to have the advantage of giving in this manner the minimum of the blood contained in the body of an adult man; for although some solid substances, not belonging to the blood, may be taken up by the water from the parenchyma of the organs permeated with capillary vessels, the excess thus obtained is so completely counteracted by the deficiency caused by the retention of some blood in the capillaries, and in part by transudation, that our estimate of the quantity of blood contained in the human body may certainly be considered as slightly below the actual quantity.

The following method, based upon the amount of sugar contained in the blood, will, I believe, afford an average estimate of the quantity of blood in animals: if we know how much sugar the blood may under the most favorable conditions contain, without its appearing in the urine, and if we determine how much sugar the blood may normally contain on an ordinary diet, we may be able to calculate the quantity of blood contained in an animal by ascertaining the quantity of sugar which must be introduced by injection into the jugular veins, or by some other method, in order to make it pass into the urine. We know from von Becker's investigations that about 0·5% of sugar may exist in the blood without passing into the urine, and that further, after the use of saccharine roots the blood contains 0·67% of sugar. Now I find from my own, as well as from Uhle and von Becker's injections of sugar (grape-sugar), that when 0·2 of a gramme of sugar was injected into the blood of rabbits of the ordinary size (1200 grammes weight), the urine indicated the presence of sugar in 25 minutes. If now we assume that in a rabbit weighing 1 kilogramme, 0·15 of a gramme of sugar injected into the blood will saturate it to such an extent, that if there were any additional quantity of sugar it would appear in the urine, a rabbit of this weight will contain 95·8 grammes of blood. Dr. von Becker is still engaged on experiments of this kind. In the present uncertainty of all the methods for determining the amount of blood in the animal body, a method like the above should not be wholly overlooked, although it may not present any great guarantee for its accuracy; since the agreement of the results of different methods would increase the degree of probability for the determination of the definite amount of blood contained in individual organisms.



It is by no means decided whether fat men and animals contain less blood than lean ones, notwithstanding the experiments of Schultz<sup>1</sup> on fat and lean oxen (in the latter, he found an excess of 20 or 30 pounds of blood). When we enter upon the consideration of the animal processes, and especially the metamorphosis of matter, we shall treat in detail of the sources from which the blood flows, its progressive and regressive formation, both in relation to its individual constituents and collectively, and of its general physiological import; for the blood is the centre round which the general metamorphosis of animal matter revolves, and in which it is perfected. As we have already considered the origin and metamorphosis of the chemical constituents of the blood, in this volume, it only remains for us here briefly to notice the mode of development and the destination of its morphological elements, although these questions may be regarded as belonging more especially to histological physiology.

The investigations of the most distinguished physiologists of the present day render it highly probable that there is more than one *seat of formation of the colorless blood-cells*. They are, undoubtedly, for the most part, formed in the chyle, and they are likewise produced, as has been before observed, in the liver; at all events, under certain conditions: but their formation, or at all events their development and growth, are not confined to any one definite locality, but proceed in the vessels of very different organs. H. Müller<sup>2</sup> and Kölliker<sup>3</sup> have recently devoted special attention to the development of the colorless blood-cells in the chyle,—a subject that had already been very fully considered by several earlier observers, especially J. Müller, E. H. Weber, Schwann, Henle, and Reichert. We find that the chyle contains numerous morphological elements, whose supposed significance as embryonic blood-corpuscles, and whose different forms in the course of their development, have led physiologists to very different views. H. Müller, who is opposed to the cell-theory of Schleiden and Schwann, thinks that the origin of these bodies from the chyle-plasma may, according to his observations, be explained somewhat in the following manner:—In the minutest lacteals there appear minute clots (solid corpuscles without a distinct cell-membrane), which are separated from the chyle, and occur as dense granules, with a viscid matter connecting them together. From these minute clots, the rudiments of the cell-wall and the nucleus are developed by a certain alteration in the chemical substrata. The nucleus appears most granular in the more recent formations, since it has been formed by the conglomeration of the insoluble and denser granules, whilst the cell-wall was being condensed into a membranous capsule. Since even at the termination of the thoracic duct we meet with minute clots in which cell-formation is only commencing, it is not improbable that their conversion into true cells—that is to say, into colorless blood-cells—is effected within the blood itself; in like manner, the first tendency towards the formation of such cells may also take place within the blood from its plasma. Müller draws attention to the fact, that most of the colorless cells of the blood contain tripartite nuclei, resembling also in this respect pus-corpuscles.

<sup>1</sup> System der Circulation. Stuttgart, 1836.

<sup>2</sup> Zeitschr. f. rat. Med. Bd. 3, S. 204-278.

<sup>3</sup> Ibid. Vol. 4, pp. 142-144.

We also find that the blood always contains cells with a simple nucleus (like the mucus-corpuscles of healthy mucous membranes); and on the other hand, that the chyle contains cells with a multiple nucleus. A slight difference in the chemical constitution of the chyle-plasma on the one side, and of the blood- or exudation-plasma in (suppuration) on the other, may perhaps be the cause of a simple nucleus in the former, and of a fissured or multiple nucleus in the latter. Kölliker strongly opposes Müller's views, and is of opinion that Schwann's theory is strictly applicable to the development of the colorless blood-corpuscles. He found at the commencement of the lacteals, but never in the thoracic duct, nuclei which were either free or surrounded by granules, and young cells with walls which almost touched the nucleus, and were very fragile. He most distinctly maintains the existence of nucleoli. Besides this origin of the lymph-corpuscles in the minutest lacteals, Kölliker also assumes that they are further augmented in the intermediate vessels, although he leaves it undecided whether this increase is effected by endogenous formation or by subdivision. The same observer distinguishes larger and smaller lymph-granules in the thoracic duct, and is of opinion that the latter only are converted into blood-corpuscles, whilst the larger gradually dissolve in the blood.

The view that the blood-cells of the embryo originate in the liver, was long since advocated by Reichert,<sup>1</sup> and recently by several physiologists, and more especially by E. H. Weber<sup>2</sup> and Kölliker.<sup>3</sup> Weber showed that in the spring the liver of frogs assumes a totally different color, while at the same season this organ is the seat of an active formation of new blood-cells. Gerlach,<sup>4</sup> whose observations have been supported by those of Schaffner,<sup>5</sup> has, however, very recently endeavored to prove that the spleen is the chief factory for the blood-cells; but the admirable chemical investigations of Scherer seem far more to corroborate the view opposed by Kölliker, and subsequently by Ecker,<sup>6</sup> that the blood-corpuscles are for the most part destroyed in the spleen. This much, at all events, seems certain, that the formation of the blood-corpuscles is not limited to definite organs, for blood-corpuscles appear in the germinal area of the embryo before the formation of vessels and glands. In the area vasculosa, blood-corpuscles and vessels are formed from cells which, according to Reichert, can in no way be distinguished from one another. There can be no doubt, therefore, that the colored blood-cells may proceed from the colorless ones; but, as yet, it remains undetermined whether such is always the order of formation, and how this mode of development is effected.

If we were to regard the colorless blood-corpuscles as merely a transition stage of formation of the colored corpuscles, their significance and physiological importance would at once be defined; but however ephemeral their existence in the blood may be, we cannot wholly deny their participation in the chemical metamorphosis of matter, more especially

<sup>1</sup> *Entwicklungsleben im Wirbelthierreich*, S. 22.

<sup>2</sup> *Zeitschr. f. rat. Med.* Bd. 4, S. 160.

<sup>3</sup> *Ibid.* Vol. 4, pp. 147-159.

<sup>5</sup> *Ibid.* Vol. 7, pp. 345-354.

<sup>4</sup> *Ibid.* Vol. 7, pp. 75-88.

<sup>6</sup> *Ibid.* Vol. 6, pp. 261-265.

as many of these bodies do not appear to be converted into colored corpuscles. They are vital cells, maintaining an active interchange of matter with the blood-plasma, and cannot therefore be wholly without influence on the general composition of the blood and the metamorphosis of the animal tissues.

With reference to the chemical phenomena accompanying the morphological transition of colorless into colored cells, we only know that hæmatin is gradually\* developed within them, and hence we must content ourselves with a brief reference to the views held by recent physiologists regarding the morphological process; omitting all notice of the older hypotheses.

The once generally accepted view that the red blood-corpuscles are formed from the nuclei of the lymph- and chyle-corpuscles, by the disappearance of their walls, has found no advocates in recent times. II. Müller, on the other hand, adopts the view that the colorless cells are directly converted into the red blood-corpuscles, and believes that the small lymph-corpuscles which occur in the thoracic duct owe their origin to the loss of their fluid granular contents, and that thus the capsule approximates more closely to the nucleus, whilst all their contents disappear so entirely in the blood, that the membrane comes in contact with, and constitutes the actual investment of the nucleus. The corpuscle is then flattened in an analogous manner to the nucleus, and appears concave, while the nucleated vesicle imbibes red pigment and thus becomes formed into a perfect blood-corpuscle. The chemical behavior of the cell-wall of the corpuscles seems however opposed to this view, and there are many other reasons unfavorable to its adoption.

According to Kölliker, the most probable view is that which assumes that the smaller kind of chyle-corpuscles is converted by the disappearance of the nucleus and the absorption of pigment into the true blood-corpuscle; he advances the following grounds in support of this view: (1) The similarity of size in the smaller chyle-corpuscles of the thoracic duct and the red blood-corpuscles; (2) the perfectly identical behavior of the capsule of these chyle-corpuscles and of the wall of the blood-disks towards physical and chemical influences; (3) the faintly yellow color of these chyle-corpuscles with an entirely colorless nucleus; (4) the flattening, although in a less degree than in fully developed blood-corpuscles; and (5) the nuclei of the smaller chyle-corpuscles are entirely different from the blood-corpuscles.

To these three theories regarding the transition of colorless into colored corpuscles Gerlach has added a fourth, which is principally founded on the occurrence of cells containing blood-corpuscles in the Malpighian corpuscles of the spleen and in the liver of the embryo. According to him, the colored blood-corpuscles are formed within the colorless ones, so that the latter stand in the relation of parent-cells to the former. But as this subject belongs less to physiological chemistry than to pure histology, the present remarks must suffice until we are able to bring chemistry to our aid in explaining the progressive and regressive development of the blood-cells. \*

As yet we know very little of the manner in which the *blood-corpuscles* act in the living blood, the objects they fulfil or the results of their

chemical metamorphoses. But our deficiency in positive knowledge has here been liberally supplied by hypotheses, whose value we will briefly consider. As might be expected, the discovery of these peculiar molecules in the blood led to that false and illogical application of the word "life," which even now is not wholly banished from physical physiology. The very vagueness of the term "life" served as a cloak for everything that did not readily admit of being referred to physical or chemical agencies. The molecules of the blood were supposed to be endowed with individual vitality like the infusoria, for which they were even mistaken by some observers (Eble and Mayer), in proof of which assertion it was maintained, according to Czermak, Treviranus and Mayer, and still more recently by Emmerson and Reader, that they exhibited a spontaneous motion. Very recently, moreover, one of our most distinguished chemists has been erroneously led by his experiments to believe in a peculiar vital activity of the blood-corpuscles. Dumas could not resist advancing the assertion that the blood-corpuscles possess a certain respiratory activity which may occasionally be reduced to actual asphyxia. It will be a sufficient refutation of this view, if we mention that Dumas was led to this conclusion merely by making the well-known observation that blood-cells, when treated with neutral alkaline salts, cohered when at rest, assume a darker color and begin to be decomposed at a moderate temperature; while this alteration occurs at a later period, when the blood which has been acted on by salts is frequently shaken. Dumas thought that the access of oxygen, brought about by shaking the blood-corpuscles, caused them to retain their vitality for a longer period; but when they are shaken with nitrogen or hydrogen gas, they do not sooner become dark than when they are shaken with atmospheric air; hence it is merely the motion which retards the cohesion and further decomposition of the blood-cells. In order to avoid misconception, we would, however, observe in reference to the vitality of the blood-cells, that if by the term "life" we mean simply a group of physical and chemical agencies, having reference to morphological progressive and regressive development, vitality can no more be denied to the blood-corpuscles than to any other animal or vegetable cell.

An opinion long prevailed that the blood-corpuscles took up oxygen in the lungs and gave it off in the capillaries, the view being based upon the bright-red color of the blood in the lungs, and its darkness in the capillaries. These cells were in fact regarded as carriers of oxygen. Henle refutes this view by observing that we might, with equal justice, also term them water-carriers, since they show themselves no less capable of absorbing the smallest additional quantity of water than of taking up oxygen and carbonic acid; for they absorb water, and again give off a portion of it, in a state of vapor, in the lungs; the gases through whose assumed chemical action this function of the cells was supposed to be derived, could only exert a mechanical influence on the form, and therefore on the color of the blood-cells. This opinion derived great probability from Mulder's careful investigation of hæmatin, which was found to be perfectly indifferent to gases, and likewise from the above-named inquiries of Nasse, Henle, Scherer, and Bruch, who have shown

the influence exerted on the color of the blood by the alterations in the form of the cells. There are, moreover, two other facts which appear to render this supposed function of the blood-cells exceedingly doubtful, if not wholly untenable; in the first place, Marchand could not obtain the slightest trace of carbonic acid in blood through which oxygen had been passed after the removal of all the gases; the conversion of oxygen into carbonic acid cannot therefore take place within the cells themselves. Another observation, made by Hannover, speaks, however, still more strongly against the usually adopted view; for this observer found that chlorotic patients, whose blood is often exceedingly deficient in colored blood-cells, exhaled in like periods of time as much carbonic acid as healthy women. Hence we might be disposed to believe with Henle, that there is no intimate relation between the corpuscles and the gases of the blood, if there were not two important grounds, supported on facts admitting of only one interpretation, which are in favor of the view according to which the blood-cells possess the capacity of absorbing oxygen. The first of these grounds rests upon the observation already referred to, that neither the intercellular fluid nor the serum alone has the power of absorbing more than a small quantity of oxygen, while the cell-containing blood exhibits a very strongly marked capacity for absorption: a fact that speaks so strongly in favor of the function of the blood-cells, that it requires no further exposition. The second ground supporting the idea of the capacity of the blood-cells for absorbing gases, is that diluted hæmatin or copiously watered blood which contains only some few recognizable corpuscles, whose contents (the hæmatin, &c.) are for the most part in a state of solution, is still susceptible to the action of carbonic acid and oxygen; the alteration of color cannot possibly depend in this case on alterations in the form of the blood-corpuscles. The hæmatin of Lecanu and Mulder is not the same as that contained in the fresh blood-cells; although the solution of the blood-cells very probably does not play the part ascribed to it, the recently dissolved hæmatin must yet participate with the blood-corpuscles in the capacity for absorbing gases. Marchand's experiment simply proves that the blood-corpuscles are not capable of generating carbonic acid by their own unaided power, or when removed from the body and brought in contact with oxygen. With respect to Hannover's view, independently of the circumstance that it admits of several modes of interpretation, it by no means overthrows the opinion that the blood-cells possess this capacity; for if a person having few blood-cells exhales as much carbonic acid as another whose blood is richer in corpuscles, it does not follow from this that the production of carbonic acid directly depends upon the blood-corpuscles, but seems rather to show the very reverse. The blood-cells, in all probability, absorb most of their carbonic acid after they reach the capillaries, and they are obviously able to take up a larger quantity than they commonly convey to the venous blood: thus 80 or 100 corpuscles of chlorotic blood may absorb the same quantity of carbonic acid as that which is generally absorbed by 120 corpuscles of healthy blood in the capillaries; these 80 cells may therefore, in like manner, exhale as much carbonic acid in the lungs as

the 120. Then, moreover, the intercellular fluid exhibits a greater capacity for dissolving carbonic acid than oxygen, and it would not therefore require the co-operation of the blood-corpuscles to convey to the lungs the carbonic acid transuded into the capillaries. We therefore consider the view which ascribes to the blood-corpuscles the function of absorbing oxygen, and giving it partially off in the capillaries, not only to be uncontroverted, but to be completely proved.

The question here arises, whether the oxygen is only mechanically taken up by the blood-cells or loosely combined with them, or whether it is chemically united with some of the individual constituents, and thus directly gives rise to the formation of carbonic acid in the capillaries. Both these modes undoubtedly occur; for the greater part of the oxygen absorbed in the lungs is only mechanically taken up by the corpuscles, or is brought to the capillaries in a slightly combined form, as is clearly proved by the experiments of Magnus, Marchand, and others; but it would be very singular if the blood-cells, which are so susceptible to external influences—as, for instance, to chemical agents—and which undoubtedly manifest an active metamorphosis of matter, should remain wholly unaffected by oxygen. This is, however, by no means the case, as we learn from direct observations. We have already shown, and purpose making still more evident by a special reference to analyses, that the difference in the chemical constitution of the arterial and venous blood-corpuscles can scarcely be explained except by the assumption of a chemical action of the oxygen upon the individual organic constituents of the blood-corpuscles in the lungs. We would here only observe, that we found the mineral substances and the hæmatin augmented in the blood-corpuscles after the inspiration of oxygen, whilst the organic substances, and more especially the fats, were considerably diminished. This incontestable fact can scarcely be explained, excepting by the supposition that it is only the mineral substances and the hæmatin which increase in weight by the absorption of oxygen, whilst the organic substances, and more especially the fats, are either destroyed by oxidation, and their products of decomposition transferred to the intercellular fluid, or at all events they undergo a considerable diminution of weight by the formation of water and carbonic acid. No one, however, can seriously believe that the blood-corpuscles swim unchanged, like mechanical molecules, from the capillaries of the lesser to those of the greater circulation.

Although we are not yet able to express the function of the blood-cells in exact chemical equations, and therefore cannot comprehend their precise physiological import, we may yet, from the facts at our disposal, form some general opinion in reference to the purpose of their existence in the blood. The blood-corpuscles are cells having special contents, whose existence cannot be conceived on physical grounds without a simultaneous and continuous metamorphosis of matter. Their activity must correspond to the menstruum in which they are suspended, and to all the relations generally in which they occur in the living body. We must, *a priori*, conclude that each recent animal cell in the healthy blood is, under given relations, metamorphosed into blood-corpuscles, precisely as

we see the primary type of the animal cell, the chyle-corpuscle, converted into a blood-cell : for it is an incontrovertible proposition in physiology, that like conditions acting on like substrata, must give rise to identical results. If, however, the formation of a cell depend upon the medium surrounding it, its subsequent activity can only be developed in relation to this medium ; hence the blood-cells and the plasma must stand in a constant reciprocal relation to one another, precisely as the yeast-cells do to the fermenting mixture. It remains, however, for future inquiries to determine the metamorphoses which result from this reciprocal action. As far as we are at present able to form an opinion on this subject, we think we shall not be deviating very widely from the truth, if we regard the blood-cells as organs, that is to say, as laboratories, in which the individual constituents of the plasma are prepared for the higher function of aiding in the formation and reproduction of the tissues. As soon, however, as we attempt to specify the individual constituents of the plasma, we lose ourselves in a labyrinth of hypotheses. Thus, for instance, some observers have conjectured that fibrin was elaborated from albumen, which is *possible*, in so far as fibrin appears to be a substance ready elaborated for deposition in the tissues, but *improbable*, since we also find in some cases that the fibrin is increased in an extraordinary degree in blood which is very poor in corpuscles (chlorosis).

The blood-corpuscles, like all other cells endowed with vital activity, have a definite period of existence. This limited duration has been regarded by some philosophical inquirers as a specific property of living beings, as if every physical or chemical process must not in like manner have a definite period of duration limited by a commencement and a termination. No one can doubt that the activity of the blood-corpuscles has its boundary, and that they perish, although the mode and course of their gradual destruction yet remains a mystery. All we know is, that in our microscopico-chemical investigations, the cells of the same blood vary in the length of time during which they can resist chemical agents, and hence it is conjectured that the more easily decomposed cells, which, moreover, generally exhibit greater intensity of color, are the older, and that those which are not easily acted upon, are paler, and appear in their granular contents to present the rudiments of a nucleus, are of more recent origin. We have no certain knowledge of the length of time that an individual cell continues to exist. The observation made by Harless, that a frog's blood-corpuscles entirely disappear after nine or ten alternations of oxygen and carbonic acid, would enable us to form an average estimate of the period of their duration, if it were not that both these substances were employed in the experiment in a state of purity, whilst in the lungs the blood-corpuscles are only acted upon by atmospheric air containing about 4% of carbonic acid. We may presume from a comparison of the blood taken in repeated venesections, that the period of duration or existence of the red corpuscles is not very short ; for the circumstance that the blood continues for several days after a moderate venesection to be poor in corpuscles, and even exhibits a great deficiency of these bodies for a prolonged period after repeated venesections certainly proves that their regeneration is not effected with great rapidity.

If, however, they are slowly regenerated, as appears to be the case, judging from the copious supply of colorless cells in the circulating fluid after severe losses of blood, they cannot have a very short existence, for otherwise the number of the colored cells would not so far exceed that of the colorless corpuscles.

The question, whether the *blood-corpuscles are disintegrated at one definite spot* has not yet been decided with any certainty. It was generally supposed by the earlier observers, that the destruction of the blood-corpuscles was effected by the alternating action of oxygen and carbonic acid, as well as of different salts and other substances, this action being gradually continued throughout the whole course of the bloodvessels, and their products also undergoing a gradual solution. As the arterial blood has been found to be poorer in corpuscles than the venous, some support seemed to be afforded to the view that the older blood-cells were principally destroyed in the capillaries of the lungs by the access of oxygen; but as it has been only proved that the weight of the sum of the blood-cells is diminished, and not that their number is lessened, we are by no means compelled to assume that the blood-cells are destroyed in the arteries; and it would even appear probable, on many grounds, that the weight of each cell is diminished by respiration, but not that the whole number is lessened. There seems, however, to have been a disposition to connect the disintegration of the blood-cells with one definite locality, and Schultz more especially designated the liver as the organ in which this process was effected. F. C. Schmid's more accurate investigations of the portal blood and of the colored cells contained in it, which differ from those of other blood, appear indeed to afford a more exact foundation for this hypothesis. We have already spoken at length of the constitution of the portal blood and of its relation to the hepatic function, in the chapters on "bile" and on "the blood," and from our comparative analyses of the portal and hepatic venous blood, we are led to the conclusion that the liver ought rather to be regarded as an organ for regenerating the blood-corpuscles than as the seat of their destruction, although we will not deny that blood-corpuscles, which have usually been regarded as cells in an advanced state of development, are conveyed from the splenic to the portal vein. On the other hand, during digestion we found only normal blood-corpuscles in the blood of the portal vein. Schultz's view cannot, therefore, be received without a certain reservation. An opinion has been lately advanced by Kölliker, and still more recently by Ecker, from the histological investigation of the spleen, and more especially of the Malpighian bodies, that this organ, which was previously held to be the seat of the formation of blood, and indeed is still regarded as such by Gerlach and Schaffner, is in fact the principal seat of the solution and complete disintegration of the blood-corpuscles. While such contending views prevail among the most trustworthy histologists, we should not venture to give the preference to either of these opposite theories, if chemical analysis did not here, as in so many cases, come to the aid of histological inquiry. Scherer has made a very admirable investigation of the spleen, which has led to several important discoveries; the chief result of which is, that in the splenic juice there occur



all the most remarkable transition stages of the products of decomposition of nitrogenous and albuminous matters, and of the blood-pigment itself. It seems highly probable from this investigation that the spleen aids in the destruction of those blood-corpuscles which are no longer able to accomplish their proper functions. We will, however, defer the fuller consideration of this hypothesis, which results from the simplest induction, till we treat of the chemico-physiological nature of the spleen,—having, moreover, already far exceeded the limits originally prescribed to the present chapter on the blood.

END OF VOL. I.





